DRAFT FOR PUBLIC COMMENTS

ÉBAUCHE POUR COMMENTAIRES DU PUBLIC

Canadian Environmental Protection Act, 1999

PRIORITY SUBSTANCES LIST

ASSESSMENT REPORT

HEXACHLOROBUTADIENE

Environment Canada
Health Canada

June 2000

TABLE OF CONTENTS

SYNOPSIS 7

1.0 INTRODUCTION 10

2.0 SUMMARY OF INFORMATION CRITICAL TO ASSESSMENT OF "TOXIC" UNDER CEPA 1999 14

2.1 IDENTITY AND PHYSICAL/CHEMICAL PROPERTIES 14
2.2 ENTRY CHARACTERIZATION 14
2.2.1 Production, importation and use *14*
2.2.2 Sources and releases *15*
2.2.2.1 Natural sources 15
2.2.2.2 Anthropogenic sources 15
2.3 EXPOSURE CHARACTERIZATION 16
2.3.1 Environmental fate *16*
2.3.1.1 Air 16
2.3.1.2 Water 16
2.3.1.3 Sediments 16
2.3.1.4 Soils 16
2.3.1.5 Biota 17
2.3.1.6 Environmental partitioning 18
2.3.2 Environmental concentrations *18*
2.3.2.1 Air 18
2.3.2.2 Drinking water 19
2.3.2.3 Surface water 19
2.3.2.4 Sediments 19
2.3.2.5 Soils 19
2.3.2.6 Biota 20
2.3.2.7 Food 20
2.3.2.8 Multimedia exposure study 20
2.4 EFFECTS CHARACTERIZATION 20
2.4.1 Ecotoxicology *20*
2.4.1.1 Pelagic organisms 20
2.4.1.2 Benthic organisms 21
2.4.2 Abiotic atmospheric effects *22*
2.4.3 Experimental animals and in vitro *23*
2.4.3.1 Acute toxicity 23
2.4.3.2 Short-term and subchronic toxicity 24
2.4.3.3 Chronic toxicity and carcinogenicity 26
2.4.3.4 Genotoxicity 26
2.4.3.5 Reproductive and developmental toxicity 27
2.4.3.6 Neurological effects and effects on the immune system 27
2.4.3.7 Toxicokinetics and mechanism of action 28
2.4.4 Humans *28*

3.0 PROPOSED ASSESSMENT OF "TOXIC" UNDER CEPA 1999 ..... **29**

3.1 CEPA 1999 64(a): ENVIRONMENT 29
3.1.1 Assessment endpoints *29*
3.1.2 Environmental risk characterization *30*
3.1.2.1 Pelagic organisms 30
3.1.2.2 Benthic organisms 31
3.1.2.3 Sources of uncertainty 33

3.1.2.4 Conclusion 33
3.2 CEPA 1999 64(b): Environment upon which life depends 33
3.3 CEPA 1999 64(c): Human health 34
3.3.1 Estimated population exposure *34*
3.3.2 Hazard characterization *35*
3.3.3 Dose–response analyses *36*
3.3.4 Human health risk characterization *38*
3.3.5 Uncertainties and degree of confidence in human health risk characterization *39*
3.4 Proposed conclusions 40
3.5 Considerations for follow-up (further action) 40

4.0 REFERENCES **47**

APPENDIX A: SEARCH STRATEGIES EMPLOYED FOR IDENTIFICATION OF RELEVANT DATA 59

LIST OF TABLES

Table 1: Risk quotient for pelagic organisms 42
Table 2: Summary of risk quotients for freshwater benthic organisms 43
Table 3: Estimated exposure of the general population to HCBD 44
Table 4: Point versus probabilistic estimates of exposure to HCBD via inhalation 45
Table 5: Critical studies and effect levels for renal toxicity in experimental animals exposed to HCBD via ingestion 46

LIST OF FIGURES

Figure 1: Structure of HCBD 14
Figure 2: Cumulative density function for HCBD in St. Clair River sediments (0–5 cm) 32

LIST OF ACRONYMS AND ABBREVIATIONS

BCF	bioconcentration factor
BMD	benchmark dose
BMD_{05}	dose associated with a 5% increase in the benchmark endpoint
$BMDL_{05}$	95% lower confidence limit on the BMD_{05}
CAS	Chemical Abstracts Service
CEPA	*Canadian Environmental Protection Act*
CEPA 1999	*Canadian Environmental Protection Act, 1999*
CFC	chlorofluorocarbon
CTV	Critical Toxicity Value
EEV	Estimated Exposure Value
ENEV	Estimated No-Effects Value
GSH	glutathione
GWP	Global Warming Potential
HCBD	hexachlorobutadiene
K_{oc}	organic carbon/water partition coefficient
K_{ow}	octanol/water partition coefficient
kg-bw	kilogram body weight
LC_{50}	median lethal concentration
LD_{50}	median lethal dose
LOAEL	Lowest-Observed-Adverse-Effect Level
LOEC	Lowest-Observed-Effect Concentration
LOEL	Lowest-Observed-Effect Level
NOAEL	No-Observed-Adverse-Effect Level
NOEL	No-Observed-Effect Level
ODP	Ozone Depletion Potential
POCP	Photochemical Ozone Creation Potential
PSL	Priority Substances List
PSL2	second Priority Substances List
TC	Tolerable Concentration
TI	Tolerable Intake

Synopsis

Hexachlorobutadiene, or HCBD, has never been commercially produced in Canada. Formerly, the substance was imported into Canada for use as a solvent, but it is no longer imported. There are no natural sources of HCBD in the environment. Current Canadian sources are minor but potentially numerous and include possible releases in landfill leachates, releases during refuse combustion and releases as a by-product in the production of some chlorinated chemicals. At present, the most significant point source of HCBD in Canada appears to be the Cole Drain, which discharges into the St. Clair River at Sarnia, Ontario, and includes outfalls from several industrial companies. The inadvertent production and use of HCBD in the United States are other potential sources of HCBD releases to the Canadian environment via long-range transport through the atmosphere or transboundary movement in shared water systems.

When released into the environment, HCBD partitions to air, soil, water and sediments, but tends to remain in the compartment to which it was released. HCBD is slowly removed from the atmosphere by photooxidation, with an estimated half-life of up to three years. Evidence for long-range transport of HCBD exists, as the substance has been detected in samples taken from various sediment depths in Great Slave Lake. HCBD biodegrades slowly in aerobic water, with an estimated half-life of up to a year, but it would persist considerably longer under anaerobic conditions. HCBD accumulates in the tissues of freshwater organisms, with a maximum reported bioconcentration factor of 19 000, but it is quite easily metabolized and therefore does not biomagnify through food chains. Available data indicate that HCBD meets the criteria for persistence and bioaccumulation according to the Persistence and Bioaccumulation Regulations of the *Canadian Environmental Protection Act, 1999* (CEPA 1999).

HCBD has been detected in Canadian surface waters, sediments, aquatic organisms and, occasionally, air.

Acute and chronic toxicity data are available for pelagic aquatic organisms, but no information is available on the toxicity of HCBD to benthic organisms.

Concentrations of HCBD in Canadian surface water are lower than the adverse effects thresholds predicted for sensitive pelagic aquatic organisms. Concentrations of HCBD in the sediment of highly contaminated sections of the St. Clair River are high enough that sensitive benthic organisms could experience adverse effects because of their inability to move to less contaminated areas.

HCBD is not likely to contribute significantly to ground-level ozone formation, but it does have the potential to contribute somewhat to depletion of stratospheric ozone and to climate change. The magnitude of these effects would depend upon the concentration of HCBD in the atmosphere; in recent years, the concentration of HCBD in Canadian air has been very low.

Available data upon which to base estimates of population exposure to HCBD in Canada are extremely limited; however, food and, possibly, air appear to be the major routes of exposure. Based on results of studies conducted in experimental animals, the kidney appears to be the target organ of HCBD-induced toxicity. Kidney tumours have also been observed in rats following long-term exposure to HCBD, but only at doses associated with non-neoplastic renal effects. The estimated average daily intake by the general population in Canada from environmental sources is less than a Tolerable Intake derived on the basis of a benchmark dose or effect levels for non-neoplastic renal effects. A Tolerable Intake is the level of intake to which it is believed a person may be exposed daily over a lifetime without deleterious effect.

Based on available data, it is concluded that hexachlorobutadiene is entering the environment in a quantity or concentration or under conditions that have or may have an immediate or long-term harmful effect on the environment or its biological diversity. It is concluded that hexachlorobutadiene is not entering the environment, in Canada, in a quantity or concentration or under conditions that constitute or may constitute a danger to the environment on which life depends; or that constitute or that may constitute a danger in Canada to human life or health. Thus, it is proposed that hexachlorobutadiene be considered "toxic" as defined in Section 64 of the *Canadian Environmental Protection Act, 1999* (CEPA 1999). Because HCBD meets the criteria for persistence and bioaccumulation according to the Persistence and Bioaccumulation Regulations of the *Canadian Environmental Protection Act, 1999* and is present in the environment as a result of human activity, the substance will be proposed, under Subsection 77(4) as a candidate for virtual elimination under Subsection 65(3).

Pursuant to subsection 77(4), because HCBD is proposed to be toxic under the Act and meets the criteria for persistence and bioaccumulation in accordance with the *Persistence and Bioaccumulation Regulations*, is present in the environment primarily as a result of human activity, and is not a naturally-occurring radionuclide or a naturally-occurring inorganic substance, implementation of virtual elimination under subsection 65(3) of HCBD is being proposed.

It is recommended that sources of HCBD as a by-product in the production of other chlorinated chemicals, such as vinyl chloride, allyl chloride and epichlorohydrin be identified and measures to reduce these releases be investigated.

HCBD releases during refuse combustion were identified. Preliminary information indicates that sources of HCBD from combustion are similar to those of dioxins, furans and hexachlorobenzene. It is recommended that measures to reduce emission of HCBD from combustion sources complement initiatives currently underway to address dioxins, furans and hexachlorobenzene.

Since HCBD is persistent, bioaccumulative, likely to cause effects to aquatic species at low levels of exposure and is not currently used in commerce in Canada, options to prevent its reintroduction into the Canadian market should be explored.

1.0 INTRODUCTION

The *Canadian Environmental Protection Act, 1999* (CEPA 1999) requires the federal Ministers of the Environment and of Health to prepare and publish a Priority Substances List (PSL) that identifies substances, including chemicals, groups of chemicals, effluents and wastes, that may be harmful to the environment or constitute a danger to human health. The Act also requires both Ministers to assess these substances and determine whether they are "toxic" or are capable of becoming "toxic" as defined in Section 64 of the Act, which states:

> ...a substance is toxic if it is entering or may enter the environment in a quantity or concentration or under conditions that
> (a) have or may have an immediate or long-term harmful effect on the environment or its biological diversity;
> (b) constitute or may constitute a danger to the environment on which life depends; or
> (c) constitute or may constitute a danger in Canada to human life or health.

Substances that are assessed as "toxic" as defined under Section 64 may be placed on Schedule I of the Act and considered for possible risk management measures, such as regulations, guidelines, pollution prevention plans or codes of practice to control any aspect of their life cycle, from the research and development stage through manufacture, use, storage, transport and ultimate disposal. Substances on Schedule I that are persistent and bioaccumulative in accordance with the Persistence and Bioaccumulation Regulations of the *Canadian Environmental Protection Act, 1999,* are present in the environment primarily as a result of human activity and are not naturally occurring radionuclides or naturally occurring inorganic substances must be proposed, under Subsection 77(4) for the implementation of virtual elimination under Subsection 65(3).

Based on initial screening of readily accessible information, the rationale for assessing hexachlorobutadiene (HCBD) provided by the Ministers' Expert Advisory Panel on the Second Priority Substances List (Ministers' Expert Advisory Panel, 1995) was as follows:

> HCBD is used as a solvent for elastomers, as a heat transfer liquid, in transformer and hydraulic fluids, and as a wash for removing volatile organic chemicals from organic streams. HCBD has been found in refuse combustion emissions and in process effluents from various industrial sectors. HCBD is highly persistent and bioaccumulative, and appears to meet the criteria of the recently adopted federal policy on toxic substances management. It is moderately to highly toxic to aquatic organisms. HCBD is carcinogenic and genotoxic in experimental animals. Potential intakes from food, based on early studies from other countries, may be close to levels that produce effects in animal studies. An assessment of the presence of HCBD in the Canadian environment is required to evaluate its potential impact on ecosystems and human health. The Panel is of the opinion that this substance should be assessed as quickly as possible.

Descriptions of the approaches to assessment of the effects of Priority Substances on the environment and human health are available in published companion documents. The document entitled "Environmental Assessments of Priority Substances under the *Canadian Environmental Protection Act*. Guidance Manual Version 1.0 — March 1997" (Environment Canada, 1997a) has

been published to provide guidance for conducting environmental assessments of Priority Substances in Canada. This document may be purchased from:

Environmental Protection Publications
Environmental Technology Advancement Directorate
Environment Canada
Ottawa, Ontario
K1A 0H3

It is also available on the Commercial Chemicals Evaluation Branch web site at www.ec.gc.ca/cceb1/eng/psap.htm under the heading "Technical Guidance Manual." It should be noted that the approach outlined therein has evolved to incorporate recent developments in risk assessment methodology, which will be addressed in future releases of the guidance manual for environmental assessments of Priority Substances.

The approach to assessment of effects on human health is outlined in the following publication of the Environmental Health Directorate of Health Canada: "*Canadian Environmental Protection Act* — Human Health Risk Assessment for Priority Substances" (Health Canada, 1994), copies of which are available from:

Environmental Health Centre
Room 104
Health Canada
Tunney's Pasture
Ottawa, Ontario
K1A 0L2

or on the Environmental Health Directorate publications web site (www.hc-sc.gc.ca/ehp/ehd/catalogue/bch.htm). The approach is also described in an article published in the *Journal of Environmental Science and Health — Environmental Carcinogenesis & Ecotoxicology Reviews* (Meek *et al.*, 1994). It should be noted that the approach outlined therein has evolved to incorporate recent developments in risk assessment methodology, which are described on the Environmental Substances Division web site (www.hc-sc.gc.ca/ehp/ehd/bch/env_contaminants/psap/psap.htm) and which will be addressed in future releases of the approach paper for the assessment of effects on human health.

The search strategies employed in the identification of data relevant to assessment of potential effects on the environment (prior to November 1997) and human health (prior to December 1996 for toxicity information) are presented in Appendix A. Review articles were consulted where appropriate. However, all original studies that form the basis for determining whether HCBD is "toxic" under CEPA 1999 have been critically evaluated by staff of Environment Canada (entry and environmental exposure and effects) and Health Canada (human

exposure and effects on human health).

The environmental sections of this Assessment Report were produced by K. Taylor, Environment Canada, based on a report entitled "*Canadian Environmental Protection Act* Environmental Assessment for Hexachlorobutadiene," which was prepared for Environment Canada under contract by P.Y. Caux and D. Moore, The Cadmus Group Inc., Ottawa, Ontario. This report was peer reviewed by:

K. Kaiser, National Water Research Institute, Environment Canada
P. Kauss, Ontario Ministry of Environment and Energy
L. McCarty, L.S. McCarty Scientific Research & Consulting

The health-related sections of this Assessment Report and supporting documentation were prepared by the following staff of Health Canada:

R. Beauchamp
K. Hughes
B. Idris
M.E. Meek

Sections of the Assessment Report and supporting documentation on genotoxicity were reviewed by D. Blakey of the Environmental Health Directorate of Health Canada. Sections related to evaluation of the effects on human health were externally reviewed by staff of BIBRA International and a peer review panel convened by Toxicology Excellence in Risk Assessment (TERA), composed of:

J. Christopher, California Environmental Protection Agency
M. Dourson, TERA
M. Friedman, Cytec Industries, Inc.
M. Gargas, ChemRisk Division of MacLaren/Hart
P. McGinnis, Syracuse Research Corporation
E. Ohanian, U.S. Environmental Protection Agency
J. Reid, University of Cincinnati

The health-related sections of the Assessment Report were reviewed and approved by the Health Protection Branch Risk Management meeting of Health Canada.

The entire Assessment Report was reviewed and approved by the Environment Canada/Health Canada CEPA Management Committee.

The text of the Assessment Report has been structured to address environmental effects initially (relevant to determination of "toxic" under Paragraphs 64(a) and (b)), followed by effects

on human health (relevant to determination of "toxic" under Paragraph 64(c)).

Copies of this Assessment Report are available upon request from:

Inquiry Centre
Environment Canada
Main Floor, Place Vincent Massey
351 St. Joseph Blvd.
Hull, Quebec
K1A 0H3

or on the Internet at:

www.ec.gc.ca/cceb1/eng/public/index_e.html

Unpublished supporting documentation, which presents additional information, is available upon request from:

Commercial Chemicals Evaluation Branch
Environment Canada
14th Floor, Place Vincent Massey
351 St. Joseph Blvd.
Hull, Quebec
K1A 0H3

or

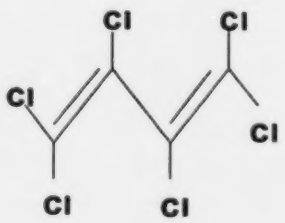
Environmental Health Centre
Room 104
Health Canada
Tunney's Pasture
Ottawa, Ontario
K1A 0L2

2.0 SUMMARY OF INFORMATION CRITICAL TO ASSESSMENT OF "TOXIC" UNDER CEPA 1999

2.1 Identity and physical/chemical properties

Hexachlorobutadiene (CAS registry number 87-68-3), referred to hereafter as HCBD, has the empirical molecular formula C_4Cl_6, the structural formula shown in Figure 1 and a molecular weight of 260.76 g/mol. HCBD is a colourless liquid with a water solubility of 3.20 mg/L at 25°C (Gradiski *et al.*, 1975), a log K_{ow} of 4.90 (Chiou, 1985), a vapour pressure of 20 Pa at 20°C (Pearson and McConnell, 1975) and a Henry's law constant of 1044 $Pa \cdot m^3/mol$ (Shen, 1982). Synonyms for HCBD include 1,1,2,3,4,4-hexachloro-1,3-butadiene, hexachloro-1,3-butadiene, perchlorobutadiene and perchloro-1,3-butadiene. Additional information on physical and chemical properties of HCBD is presented in Environment Canada (1999).

Figure 1: Structure of HCBD



2.2 Entry characterization

2.2.1 Production, importation and use

HCBD has never been commercially produced in Canada. It is produced as a by-product during the production of certain chlorinated chemicals, such as tetrachloroethylene, trichloroethylene, vinyl chloride, allyl chloride, epichlorohydrin and carbon tetrachloride (U.S. EPA, 1980; Kusz *et al.*, 1984; Choudhary, 1995).

In the past, HCBD was imported into Canada for use as a solvent (Environment Canada, 1979), but it is no longer imported or used (Environment Canada, 1997c). In addition, HCBD was not included on the National Pollutant Release Inventory (NPRI, 1994).

HCBD was used as a solvent for C_4 and higher hydrocarbons and elastomers, as a hydraulic fluid, as a heat transfer liquid in transformers and as a chemical intermediate in the production of chlorofluorocarbons and lubricants (U.S. EPA, 1980; Manahan, 1992). It was also

used to recover chlorine-containing gas in chlorine plants, in gyroscopes and in insulating fluids, and it had widespread application as a fumigant for treating grapes against *Phylloxera* in the former Soviet Union, France, Italy, Greece, Spain and Argentina (IARC, 1979; IPCS, 1994). Recent information on the use of HCBD is not available (IPCS, 1994).

2.2.2 Sources and releases

2.2.2.1 Natural sources

There are no natural sources of HCBD in the environment.

2.2.2.2 Anthropogenic sources

In the 1970s, formation of HCBD as a waste by-product was estimated to be 1.5% of total tetrachloroethylene production (Brown *et al.*, 1975). Some of this waste was emitted to the aquatic environment in industrial effluents and to air from stacks. Since the closing of the two tetrachloroethylene plants in Canada in 1985 and 1992, there have been no major point sources of HCBD. Current Canadian sources are minor but potentially numerous. They include possible releases in landfill leachates, releases during refuse combustion and releases as a by-product in the production of other chlorinated chemicals, such as vinyl chloride, allyl chloride and epichlorohydrin.

Based on 12-month average concentrations, HCBD was detected (detection limit 10 ng/L) in 4 of 26 effluent streams from organic chemical manufacturing plants in Ontario and in 9 of 74 final discharge streams monitored between 1989 and 1991. Estimated loadings at these sites ranged from <1 to 9 g/day; the total loading from this sector was estimated to be 20 g/day (OME, 1992). The most significant point source of HCBD appears to be the Cole Drain, which discharges into the St. Clair River at Sarnia, Ontario, and includes outfalls from several industrial companies. Loadings from the Cole Drain appear to have decreased from 140 g/day in 1985 (OME, 1991) to 30 g/day in 1995 (Kauss, 1996). In a survey of the Cole Drain final mixing chamber discharge in 1995, a maximum concentration of 0.9 µg HCBD/L was detected (Kauss, 1996).

The inadvertent production and use of HCBD in the United States are other potential sources of HCBD releases to the Canadian environment through atmospheric long-range transport or transboundary movement in shared water systems. Evidence for long-range transport of HCBD was provided by Mudroch *et al.* (1992), who found that HCBD was present at concentrations ranging from 0.01 to 0.23 ng/g at various sediment depths in samples taken from Great Slave Lake in 1987. According to the United States Toxic Release Inventory, 2 tonnes of HCBD were released to the environment in the United States in 1995; 75% of this total was to the air, 15% to water and 10% to underground injection (Toxic Release Inventory, 1997). The load to the atmosphere, however, does not include all possible releases from every type of

industrial facility (ATSDR, 1994).

2.3 Exposure characterization

2.3.1 Environmental fate

2.3.1.1 Air

In air, HCBD persists until it is either degraded photochemically or adsorbed to particulate matter and deposited to water or soil. Estimates of its half-life in air based on photochemical degradation through reactions with hydroxyl radicals and ozone range from 60 days (ATSDR, 1994) to three years (Howard *et al.*, 1991).

Class and Ballschmiter (1987) calculated that HCBD would have a tropospheric half-life of 840 days in the northern hemisphere and 290 days in the southern hemisphere, based on a hydroxyl radical rate constant of 2×10^{-14} cm^3/molecule per second and a hydroxyl radical concentration of 7×10^5 molecules/cm^3 in the north and 17×10^5 molecules/cm^3 in the south.

These data indicate that HCBD meets the criteria for persistence in air (half-life ≥2 days) in accordance with the Persistence and Bioaccumulation Regulations of the *Canadian Environmental Protection Act, 1999*.

2.3.1.2 Water

HCBD was completely degraded by wastewater microbiota within seven days of exposure under aerobic conditions (Tabak *et al.*, 1981). Degradation of HCBD is very slow under anaerobic conditions (Johnson and Young, 1983; Govind *et al.*, 1991; Howard, 1991). The half-life of HCBD in water is proportional to the amount of organic matter in the aqueous media; in natural waters, the half-life is estimated to be 4–52 weeks (Howard *et al.*, 1991).

2.3.1.3 Sediments

Sediments are a sink for HCBD released to water. In sediments with high organic content, the compound is not expected to persist; however, measured values for the half-life in sediment are not available. HCBD will eventually be biodegraded in aerobic sediments.

2.3.1.4 Soils

The half-life of HCBD in soil depends upon the chemical, physical and biological heterogeneity of the soil and climatic conditions. Howard *et al.* (1991) estimated the half-life to be 4–26 weeks, based on aerobic biodegradation rates; these authors suggested that HCBD may not biodegrade in anaerobic zones of soil and that evaporation would be a significant transport mechanism from soil

surfaces. In a dune infiltration study in the Netherlands, HCBD was found to be mobile in sandy soils, with an average residence time of 100 days and little biodegradation (Howard, 1991).

Fragiadakis *et al.* (1979) examined residues of radio-labelled HCBD in soil–plant systems and observed that 4% of the original radioactivity was bound in non-extractable residues in the top 50 cm of soil after two years, suggestive of potential long-term accumulation. The remaining 96% of the original radioactivity was unaccounted for and was believed to have volatilized.

2.3.1.5 Biota

HCBD partitions preferentially into lipid phases. Although HCBD accumulates in the tissues of freshwater aquatic invertebrates and fish, it does not biomagnify through food chains because of its fast depuration rate (Environment Canada, 1983). HCBD tends to be preferentially accumulated in the livers of fish. The bioconcentration factors (BCFs) in muscle and liver were 700 and 10 000, respectively, in dab, *Limanda limanda* (Pearson and McConnell, 1975). HCBD was eliminated from the tissues of goldfish (*Carassius auratus*) with a half-life of 6.3 days (Leeuwangh *et al.*, 1975).

BCFs ranging from 1 to 19 000 on a whole-body basis have been reported for HCBD in the literature. The highest BCF reported was determined in rainbow trout (*Oncorhynchus mykiss*) in a field study (Oliver and Niimi, 1983). This wide range can be explained in part by species differences in metabolism or differences in exposure concentrations (ATSDR, 1994). It takes longer for equilibrium to be reached in fish at lower exposure concentrations than at higher levels (69 days at 0.1 ng/L versus 7 days at 3.4 ng/L) (Oliver and Niimi, 1983). BCFs were more than two-fold greater at the higher exposure levels than at the lower concentrations, indicating that rates of detoxification and elimination by fish are concentration dependent.

HCBD also bioconcentrates in aquatic invertebrates, but to a somewhat lesser degree than in fish, with a maximum reported BCF of 2000 for the mussel, *Mytilus edulis* (Pearson and McConnell, 1975). Contamination of water by HCBD led to uptake of the substance by caged mussels in the St. Clair River (Kauss and Hamdy, 1985; OME/MDNR, 1991).

HCBD does not appear to bioaccumulate in plants. In a field study with radio-labelled HCBD, no significant degree of accumulation occurred in roots, leaves or stems of potato or carrot plants (Fragiadakis *et al.*, 1979).

The available data for fish indicate that HCBD meets the criteria for bioaccumulation (BCF ≥5000) in accordance with the Persistence and Bioaccumulation Regulations of the *Canadian Environmental Protection Act, 1999*.

2.3.1.6 Environmental partitioning

The distribution of HCBD in the environment was estimated using EQC Level III, a steady-state, non-equilibrium fugacity model (DMER and AEL, 1996). The results of the modelling show that HCBD tends to remain in the environmental compartment into which it is released. If HCBD is emitted into air, more than 98% would be found in the air, about 1% in soil and less than 1% in water and sediments. If released to soil, about 99% would be found in the soil and about 1% in air. If released to water, about 70% would be found in the water, about 15% in each of air and sediments and less than 1% in soil. Values for input parameters were as follows: molecular weight, 260.76 g/mol; vapour pressure, 20 Pa; water solubility, 3.20 mg/L; log K_{ow}, 4.90; Henry's law constant, 1044 Pa·m^3/mol; half-life in air, 1700 hours; half-life in water, 550 hours; half-life in soil, 550 hours; and half-life in sediment, 550 hours. Justification for the selection of these input parameters is presented in DMER and AEL (1996). Modelling was based on an assumed default emission rate of 1000 kg/hour into a region of 100 000 km^2, which includes a surface water area (20 m deep) of 10 000 km^2. The height of the atmosphere was assumed to be 1000 m. Sediments and soils were assumed to have an organic carbon content of 4% and 2% and a depth of 1 cm and 10 cm, respectively. The estimated percent distribution predicted by this model is not affected by the assumed emission rate.

The predicted distributions suggest that little intermedia transport will occur when HCBD is discharged to air or soil. By comparison, disposal to water has the potential for significant transport of HCBD to the air and sediment compartments.

2.3.2 Environmental concentrations

The closure of tetrachloroethylene production plants, changes in industrial processes and improvements in waste treatment processes, including improvements in containment facilities and spill prevention, have resulted in greatly reduced loadings of HCBD in the Canadian environment since the early 1980s; HCBD has only rarely been detected in recent monitoring programs in areas removed from former sources.

2.3.2.1 Air

HCBD was detected (detection limit 0.1 µg/m^3) in only 153 of 9231 samples (i.e., less than 2%) of outdoor air from 46 sites across Canada surveyed from 1989 to early 1997. It has not been detected at any of these sites since 1994. The maximum concentration measured was about 4 µg/m^3 in Windsor in 1992. Mean concentrations at each site, calculated by assuming a concentration of one-half the detection limit of 0.1 µg/m^3 in those samples that did not contain detectable levels of HCBD, ranged from 0.05 to 0.07 µg/m^3 (Dann, 1997).

No data on levels of HCBD in indoor air in Canada or in "uncontaminated" areas in other countries were identified.

2.3.2.2 Drinking water

HCBD has not been detected in drinking water (detection limits ranging from 0.7 pg/L to 5 µg/L) in most provincial monitoring programs in Canada (Environment Ontario, 1987; Kendall, 1990; Jobb *et al.*, 1993; Alberta Environmental Protection, 1996; Riopel, 1996; Zanette, 1996). It was detected (detection limit 1 ng/L) in only 5 of 2994 samples of treated drinking water from 143 sites across Ontario surveyed in 1991–1995; the maximum concentration measured was 6 ng/L in Port Dover (OMEE, 1996).

2.3.2.3 Surface water

The highest reported concentration of HCBD in Canadian surface waters was 1.3 µg/L, which was measured in the St. Clair River in 1984 (OME/MDNR, 1991); levels have decreased substantially (i.e., 500-fold) since 1984, based on a measurement of 0.0027 µg/L downstream from the Cole Drain in 1994, the highest concentration reported that year (Kauss, 1996). Since 1990, concentrations of HCBD in surface water from southern Ontario have generally been less than 0.001 µg/L (Environment Canada *et al.*, 1995; L'Italien, 1996). A maximum concentration of 24 µg/g dry weight was measured in suspended sediments from the St. Clair River in 1985 (Oliver and Kaiser, 1986); in 1989, the highest level detected was 0.01 µg/g dry weight (Chan, 1993).

2.3.2.4 Sediments

The maximum level of HCBD in sediment in the St. Clair River, near Sarnia, Ontario, where the greatest contamination by HCBD in Canada has reportedly occurred, prior to 1986 was 430 µg/g dry weight (lowest reported concentration of 0.0001 µg/g dry weight); it was detected (detection limit not specified) in 59 of 65 sampling sites in 1985 (Oliver and Pugsley, 1986). The highest concentration measured in recent years was 310 µg/g dry weight, downstream from the Cole Drain at a depth of 5–15 cm in 1994; in this survey, HCBD was detected (detection limit 0.001 µg/g dry weight) in 148 of 153 samples (Farara and Burt, 1997; Kauss, 1997). In the top 5 cm of sediment in a 2-km stretch of the St. Clair River in an industrialized zone in 1994, concentrations of HCBD ranged from <0.001 to 243 µg/g dry weight (detectable in 37 of 39 samples; detection limit 0.001 µg/g dry weight), with a geometric mean of 0.64 µg/g dry weight (Bedard and Petro, 1997). In these samples, the 99th-, 95th- and 90th-percentile values were 194, 60.9 and 18.7 µg/g dry weight, respectively, while the median was 0.9 µg/g dry weight.

2.3.2.5 Soils

In the only identified relevant survey in Canada, HCBD was not detected (detection limit 0.05 µg/g dry weight) in 24 samples of agricultural soils from across the country or in 6 samples from areas that had repeatedly received heavy applications of pesticides (Webber and Wang, 1995).

2.3.2.6 Biota

No recent data on HCBD concentrations in biota have been identified. Levels in rainbow trout collected from Lake Ontario in 1981 ranged from 0.06 to 0.3 ng/g (mean 0.2 ng/g) (Oliver and Niimi, 1983). Levels of up to 10 ng/g have been detected in composite samples of coho salmon (*Oncorhynchus kisutch*) collected from the Great Lakes in 1980 (Clark *et al.*, 1984). The maximum concentration of HCBD in caged mussels, *Elliptio complanata* following three weeks of exposure on the sediment surface near three industrial areas of the St. Clair River was 36 ng/g wet weight (Kauss and Hamdy, 1985; OME/MDNR, 1991; Kauss, 1997).

2.3.2.7 Food

Data on levels of HCBD in foodstuffs (in addition to those discussed in Section 2.3.2.6) are limited primarily to earlier studies conducted in other countries. Concentrations of HCBD in beverages, bread, butter, cheese, eggs, fruits, meats, milk, oils and potatoes ranging from non-detectable to 3.7 µg/kg (grapes) were reported in the United Kingdom (McConnell *et al.*, 1975), while in Germany, concentrations of HCBD in chicken, eggs, fish, margarine, meat and milk ranged from non-detectable to 42 µg/kg (egg yolk) (Kotzias *et al.*, 1975) (detection limits were not specified in either report). HCBD was not detected in samples of eggs or vegetables and was detected in only 1 of 20 samples of milk produced or grown in the vicinity of organic chemical manufacturing plants in the United States (detection limits of 5 or 40 µg/kg) (Yip, 1976). In a survey of breast milk of women from five regions of Canada, HCBD was not detected in any of 210 samples analysed (detection limit 1.2 µg/L) (Mes *et al.*, 1986).

2.3.2.8 Multimedia exposure study

In a recent pilot multimedia exposure study, samples of personal air, tap water, beverages and food from 44 households in the Toronto area were analysed for HCBD. None of the samples contained detectable amounts of HCBD, although the detection limits in this study were generally higher than those reported in other studies discussed above (i.e., 0.64 µg/m^3 for air, 2 µg/L for water and 0.09–0.9 µg/kg for food and beverages), and the analytical recovery of HCBD was not determined (Zhu, 1997).

2.4 Effects characterization

2.4.1 Ecotoxicology

2.4.1.1 Pelagic organisms

HCBD preferentially accumulates in the livers of fish (Pearson and McConnell, 1975). Once in the liver, it can be biotransformed into polar metabolites that will reach the kidneys via the bile and

could become nephrotoxic in fish (Anders and Jakobson, 1985; Yang, 1988; IPCS, 1994).

The available data on toxicity for sensitive receptors indicate that chronic effects occur at concentrations an order of magnitude below those causing acute effects. In most cases, freshwater fish and marine crustacea are more sensitive than their marine and freshwater counterparts, respectively.

The lowest available chronic value was a 28-day Lowest-Observed-Effect Concentration (LOEC) of 13 µg/L reported for the fathead minnow (*Pimephales promelas*), based on survival and growth (Benoit *et al.*, 1982). No chronic data on toxicity were identified for aquatic invertebrates. The lowest identified acute value was a 96-hour LC_{50} of 32 µg/L for the marine mysid shrimp, *Mysidopsis bahia* (U.S. EPA, 1980). For fish, the lowest identified acute value was a 96-hour LC_{50} of 90 µg/L for the goldfish (Leeuwangh *et al.*, 1975). In other studies, acute toxicity was reported only at concentrations of HCBD above 100 µg/L (Pearson and McConnell, 1975; Laseter *et al.*, 1976; Dow Chemical Co., 1978; Juhnke and Lüdemann, 1978; Laska *et al.*, 1978; Slooff, 1979; U.S. EPA, 1980; Walbridge *et al.*, 1983; Geiger *et al.*, 1985; Mayer and Ellersieck, 1986; Roederer *et al.*, 1989). The most sensitive freshwater invertebrate identified was the aquatic sowbug, *Asellus aquaticus*, with a 96-hour LC_{50} of 130 µg/L (Leeuwangh *et al.*, 1975). Bacteria and plants are less sensitive to HCBD than fish or invertebrates (Knie *et al.*, 1983).

2.4.1.2 Benthic organisms

There were no acute or chronic toxicity studies using benthic organisms identified for HCBD. In the absence of such data, the water–sediment Equilibrium Partitioning approach can be used to estimate a Critical Toxicity Value (CTV) for HCBD for benthic organisms. The principle behind this approach is that sediment organic carbon is the main factor influencing partitioning of non-polar organic compounds into sediments (Di Toro *et al.*, 1991). For HCBD, the CTV for the most sensitive freshwater pelagic invertebrate multiplied by the organic carbon/water partition coefficient (K_{oc}) and the organic content of the sediment (f_{oc}) can be used to estimate a CTV for benthic organisms using the equation:

$$CTV_{benthic} = f_{oc} \times K_{oc} \times CTV_{pelagic}$$

where:

- f_{oc} is 0.02, based on the mean organic carbon content for all surficial sediment samples from the St. Clair River in 1994, expressed on a dry-weight basis (Kauss, 1997),
- K_{oc} is 80 000 L/kg, based on the log K_{oc} of 4.90 (Oliver and Kaiser, 1986) for HCBD, and
- $CTV_{pelagic}$ is 13 µg/L, the 28-day LOEC for fathead minnow (Benoit *et al.*, 1982).

Therefore:

$$CTV_{benthic} = 0.02 \times 80\,000 \text{ L/kg} \times 13 \text{ µg/L}$$

$$= 20\,800 \text{ µg/kg dry weight}$$

$$= 20.8 \text{ µg/g dry weight}$$

The CTV for HCBD for benthic organisms is therefore estimated to be 20.8 µg/g dry weight.

2.4.2 Abiotic atmospheric effects

Class and Ballschmiter (1987) calculated that HCBD would have a tropospheric half-life of 840 days in the northern hemisphere and 290 days in the southern hemisphere. These half-lives are sufficiently long to allow HCBD to reach the stratosphere and react with the ozone present there (Bunce, 1996).

Worst-case calculations were made to determine if HCBD has the potential to contribute to depletion of stratospheric ozone, ground-level ozone formation or climate change (Bunce, 1996).

The Ozone Depletion Potential (ODP) was calculated to be 0.07 (relative to the reference compound CFC-11, which has a ODP of 1), based on the following formula:

$$ODP = (t_{HCBD}/t_{CFC\text{-}11}) \times (M_{CFC\text{-}11}/M_{HCBD}) \times ([n_{Cl} + \alpha n_{Br}]/3)$$

where:

- t_{HCBD} is the lifetime of HCBD (4.2 years),
- $t_{CFC\text{-}11}$ is the lifetime of CFC-11 (60 years),
- $M_{CFC\text{-}11}$ is the molecular weight of CFC-11 (137.5 g/mol),
- M_{HCBD} is the molecular weight of HCBD (260.8 g/mol),
- n_{Cl} is the number of chlorine atoms in the HCBD molecule (6),
- n_{Br} is the number of bromine atoms in the HCBD molecule (0), and
- α is a measure of the effectiveness of bromine in ozone depletion with respect to chlorine.

The Photochemical Ozone Creation Potential (POCP) was estimated to be 0.01 (relative to the value of an equal mass of the reference compound ethene, which has a POCP of 100), based on the following formula:

$$POCP = (k_{HCBD}/k_{ethene}) \times (M_{ethene}/M_{HCBD}) \times 100$$

where:

- k_{HCBD} is the rate constant for the reaction of HCBD with OH radicals (9.5×10^{-15} cm^3/mol per second),
- k_{ethene} is the rate constant for the reaction of ethene with OH radicals (8.5×10^{-12} cm^3/mol per second),
- M_{ethene} is the molecular weight of ethene (28.1 g/mol), and
- M_{HCBD} is the molecular weight of HCBD (260.8 g/mol).

The Global Warming Potential (GWP) was calculated to be 0.037 (relative to the reference compound CFC-11, which has a GWP of 1), based on the following formula:

$$GWP = (t_{HCBD}/t_{CFC\text{-}11}) \times (M_{CFC\text{-}11}/M_{HCBD}) \times (S_{HCBD}/S_{CFC\text{-}11})$$

where:

- t_{HCBD} is the lifetime of HCBD (4.2 years),
- $t_{CFC\text{-}11}$ is the lifetime of CFC-11(60 years),
- $M_{CFC\text{-}11}$ is the molecular weight of CFC-11 (137.5 g/mol),
- M_{HCBD} is the molecular weight of HCBD (260.8 g/mol),
- S_{HCBD} is the infrared absorption strength of HCBD (2389/cm^2 per atmosphere, default), and
- $S_{CFC\text{-}11}$ is the infrared absorption strength of CFC-11 (2389/cm^2 per atmosphere).

These figures imply that HCBD is not likely to contribute significantly to ground-level ozone formation, but it does have the potential to contribute somewhat to depletion of stratospheric ozone and to climate change.

2.4.3 Experimental animals and in vitro

2.4.3.1 Acute toxicity

HCBD is moderately acutely toxic, with LD_{50}s of 65–116 mg/kg-bw in mice, 200–580 mg/kg-bw in rats and 90 mg/kg-bw in guinea pigs (Murzakaev, 1963; Gulko *et al.*, 1964; Gradiski *et al.*, 1975; Kociba *et al.*, 1977a, 1977b). Birner *et al.* (1995) observed necrosis of the pars recta of the proximal renal tubules in Wistar rats administered a single dose of 200 mg/kg-bw; renal tubular necrosis was also induced in laboratory animals exposed to single doses of several metabolites of HCBD (Lock and Ishmael, 1979; Jaffe *et al.*, 1983; Lock *et al.*, 1984; Nash *et al.*, 1984).

2.4.3.2 Short-term and subchronic toxicity

Although the database is limited, in available short-term and subchronic studies in rats and mice, the renal proximal tubules appear to be the principal site of injury at the lowest doses that cause effects following oral or inhalation exposure. Although decreases in body weight gain were sometimes also observed at the lowest exposure levels at which effects were observed, these decreases were generally associated with reduced food consumption.

Increased relative kidney weight and histopathological changes, including degeneration of the proximal tubular epithelial cells, necrosis and regeneration, and alterations in biochemical parameters in the blood and urine (consistent with renal damage) were reported in short-term studies in Wistar or Sprague-Dawley rats exposed to HCBD in the diet or by gavage for 2–4 weeks at doses as low as 2.5 mg/kg-bw per day (Kociba *et al.*, 1971; Harleman and Seinen, 1979; Stott *et al.*, 1981; Jonker *et al.*, 1993). Jonker *et al.* (1993) observed female rats to be more sensitive to the nephrotoxic effects than male rats, as histopathological changes in the kidney occurred in females at 100 and 400 ppm in the diet (approximately equivalent to doses of 5 and 20 mg/kg-bw per day, respectively) and in males only at 400 ppm, although effects on kidney weight and biochemical parameters were noted in both sexes at 100 ppm and above. In the only identified short-term study in mice, there was a dose-related increase in severity of renal toxicity, characterized by pale kidney cortices and necrosis of the cortex and/or outer medulla, in male and female $B6C3F_1$ mice administered concentrations of HCBD equivalent to doses as low as 3 mg/kg-bw per day in the diet for two weeks (Yang *et al.*, 1989; NTP, 1991).

In a subchronic study in which groups of 10 male or female Wistar-derived rats were administered doses of 0, 0.4, 1.0, 2.5, 6.3 or 15.6 mg HCBD/kg-bw per day in arachid oil by gavage for 13 weeks, there was a dose-related increase in relative kidney weight, which was significant in females at the two highest doses and at all doses in males. Histopathological changes in the kidney, consisting of large, prominent hyperchromatic nuclei and focal necrosis of epithelial cells and nuclear detritus, were observed in the renal proximal tubules in females at 2.5 mg/kg-bw per day and above and in males at 6.3 mg/kg-bw per day and above. There were also dose-related decreases in urine osmolarity (indicative of compromised urine-concentrating ability of the kidneys), which were significant in females at 2.5 mg/kg-bw per day and above and in males at the highest dose only (Harleman and Seinen, 1979). The Lowest-Observed-Adverse-Effect Levels (LOAELs), based on renal effects, are considered to be 2.5 and 6.3 mg/kg-bw per day in females and males, respectively, with No-Observed-Adverse-Effect Levels (NOAELs) of 1.0 and 2.5 mg/kg-bw per day in females and males, respectively (the authors presented these latter values as "no-effect levels").

Effects on the kidney were also observed in groups of 10–34 male or female Sprague-Dawley rats administered doses of 0, 0.2, 2.0 or 20 mg HCBD/kg-bw per day in the diet for approximately 148 days. The kidneys of only five animals per group were examined histopathologically. The relative weight of the kidney was significantly increased in both sexes at

20 mg/kg-bw per day, whereas the kidneys in males administered the two highest doses were "roughened" and mottled in appearance. There was minimal or moderate renal tubular dilation and hypertrophy with foci of renal tubular epithelial degeneration and regeneration in four of five male or female rats in the high-dose group; these lesions also occurred in one female at 2.0 mg/kg-bw per day. Renal changes that are characteristic of this strain of rats occurred in all dose groups, but with greater severity at 2.0 and 20 mg/kg-bw per day (Schwetz *et al.*, 1977). The No-Observed-Effect Level (NOEL) and Lowest-Observed-Effect Level (LOEL) for effects on the kidney are considered to be 0.2 and 2.0 mg/kg-bw per day, respectively. (Note: The latter value was not considered a LOAEL because of the lack of statistical significance of the observed effects.)

In the only subchronic study in mice, diets containing 0, 1, 3, 10, 30 or 100 ppm HCBD (which the authors calculated to be equivalent to doses of 0, 0.1, 0.4, 1.5, 4.9 and 16.8 mg/kg-bw per day for males and 0, 0.2, 0.5, 1.8, 4.5 and 19.2 mg/kg-bw per day for females) were administered to groups of 10 $B6C3F_1$ mice of each sex for 13 weeks. Dose-related reductions in relative and/or absolute kidney weights were reported; these reductions were significant in males in the three highest dose groups and in females in the two highest dose groups. The incidence and severity of renal tubular epithelial regeneration, characterized by increased basophilia of the tubular cell cytoplasm, occasional mitosis and an increased number of nuclei, increased in an exposure-related manner (0/10, 1/10, 9/10, 10/10, 10/10 and 10/10 [females] and 0/10, 0/10, 0/10, 0/9, 10/10 and 10/10 [males] at 0, 1, 3, 10, 30 and 100 ppm, respectively). Females appeared to be more sensitive than males, as the incidence of this lesion was significantly increased at 3 ppm and above in females and at 30 ppm and above in males; renal tubular regeneration was also observed in 1 of 10 female mice exposed to 1 ppm. (The lesion in this mouse was assigned a severity score of 2; Elwell, 1993.) Unlike the observation of renal necrosis in the short-term study, only regenerative changes were observed in this study, which the authors suggested was indicative of adaptation and compensation by the kidney tubular epithelium for cell loss. Based on the histopathological effects in the kidney, the authors considered the NOAEL in male mice to be 1.5 mg/kg-bw per day; a no-effect level for female mice was not presented by the authors, as renal tubular regeneration was observed in all dose groups (Yang *et al.*, 1989; NTP, 1991). Therefore, because of the lack of statistical significance of the response in the female mice in the lowest dose group (for which data on the incidence of this lesion in historical controls at the National Toxicology Program were not available for comparison) and the severity of the renal tubular regeneration in the one mouse in this dose group, as well as the lack of data on food consumption for individual animals (i.e., it is unclear whether this effect may have been a function of increased food consumption), 0.2 mg/kg-bw per day is considered to be the LOEL for renal toxicity in females in this study.

In the only short-term or subchronic study identified in which animals were exposed to HCBD by inhalation, renal proximal tubular degeneration and adrenal cortical degeneration were noted in groups of four male or female Alderley Park SPF rats exposed to 25 ppm (267 mg/m^3) HCBD and above for up to 15 days. Renal toxicity was not observed at lower concentrations (5

ppm [53 mg/m^3] or 10 ppm [107 mg/m^3]) (Gage, 1970).

2.4.3.3 Chronic toxicity and carcinogenicity

The identified information on the chronic toxicity and carcinogenicity of HCBD is extremely limited. In the only long-term study identified, groups of 39 or 40 (90 in controls) male and female Sprague-Dawley rats were administered doses of 0, 0.2, 2.0 or 20 mg HCBD/kg-bw per day in the diet for two years. Mortality was significantly increased in males in the 20 mg/kg-bw per day group during the last two months of the study. Body weight gain was significantly decreased and absolute and relative kidney weights were significantly increased in both sexes at this dose. There were significant increases in urinary coproporphyrin in males and females at 20 mg/kg-bw per day and in females at 2.0 mg/kg-bw per day; however, other urinary biochemical parameters were not altered. Histopathological changes, including multifocal or disseminated hyperplasia and focal adenomatous proliferation of the renal tubular epithelium, were observed in rats exposed to the highest dose and "possibly" at 2.0 mg/kg-bw per day, with females being more sensitive than males (incidence and statistical significance not specified). The incidence of renal tumours (adenomas, adenocarcinomas and carcinomas, combined) was significantly increased in rats of both sexes administered 20 mg/kg-bw per day (males: 1/90 [1.1%], 0/40 [0%], 0/40 [0%] and 9/39 [23.1%] at 0, 0.2, 2.0 and 20 mg/kg-bw per day, respectively; females: 0/90 [0%], 0/40 [0%], 0/40 [0%] and 6/40 [15.0%] at 0, 0.2, 2.0 and 20 mg/kg-bw per day, respectively). There were no significant increases in the incidence of tumours at other sites. The authors concluded that HCBD induced renal tumours only at a dose level greater than that which caused observable non-neoplastic injury (Kociba *et al.*, 1977a). The NOEL for non-neoplastic kidney damage was considered to be 0.2 mg/kg-bw per day, with a LO(A)EL of 2.0 mg/kg-bw per day. (It is not possible to determine whether the effects at this dose were adverse on the basis of information presented in the published account of the study.)

Additional limited screening bioassays contribute little to the assessment of the potential carcinogenicity of HCBD. HCBD did not induce local or distant tumours following chronic dermal application or short-term intraperitoneal administration in sensitive strains of mice (Theiss *et al.*, 1977; Van Duuren *et al.*, 1979), although the extent of histopathological examination was limited in these studies; nor did HCBD initiate the induction of skin papillomas in mice in a long-term initiation-promotion assay (Van Duuren *et al.*, 1979).

2.4.3.4 Genotoxicity

Although the results of available studies are not completely consistent, there is some limited evidence that HCBD is genotoxic under certain conditions. The results of early standard Ames tests were negative in both the presence and absence of liver S-9 metabolic activation (De Meester *et al.*, 1980; Stott *et al.*, 1981; Haworth *et al.*, 1983; Reichert *et al.*, 1983). However, HCBD induced gene mutations in *Salmonella typhimurium* in the presence of liver S-9 mix with enhanced protein content (Reichert *et al.*, 1984) and in the presence of liver microsomes and

glutathione (GSH), with a greater response with both liver and kidney microsomes and GSH (Vamvakas *et al.*, 1988). Positive results were also obtained for the Ara test in *Salmonella*, only in the absence of liver S-9 metabolic activation (Roldán-Arjona *et al.*, 1991). HCBD induced sister chromatid exchanges in Chinese hamster ovary cells (with and without S-9), but not chromosomal aberrations (Galloway *et al.*, 1987); chromosomal aberrations were also not induced in peripheral human lymphocytes, although the exposure levels tested were much lower (German, 1988).

One author reported the induction of chromosomal aberrations in the bone marrow of mice exposed to HCBD orally (≥2 mg/kg-bw) or by inhalation (10 mg/m^3) (German, 1988), whereas negative results have been reported in other studies in rats exposed to greater concentrations or doses (Schwetz *et al.*, 1977; NIOSH, 1981). Increased DNA synthesis and minor amounts of DNA alkylation were observed in the kidney of rats administered single or repeated oral doses of 20 mg HCBD/kg-bw (Stott *et al.*, 1981). In addition, there was significant covalent binding to mitochondrial DNA in the kidney of mice orally exposed to 30 mg HCBD/kg-bw (Schrenk and Dekant, 1989).

Several of the metabolites of HCBD have been mutagenic in *Salmonella*. The cysteine conjugate, which appears to be the most potent of the metabolites tested, is likely cleaved by bacterial β-lyase to mutagenic intermediates (Dekant *et al.*, 1986). The mutagenic activity of the S-conjugate is enhanced by the presence of rat renal microsomes and mitochondria, which exhibit high γ-glutamyl transpeptidase activity (Vamvakas *et al.*, 1988). Similarly, the mercapturic acid metabolite was mutagenic only in the presence of metabolic activation, which would provide N-deacetylase (Wild *et al.*, 1986), whereas the bis-conjugates were not active under any conditions (Vamvakas *et al.*, 1988).

2.4.3.5 Reproductive and developmental toxicity

Subchronic or chronic oral administration of up to 20 mg HCBD/kg-bw per day did not induce histopathological changes in the testes or ovaries or effects on estrous cycle or sperm parameters in B6C3F$_1$ mice or Sprague-Dawley rats (Kociba *et al.*, 1977a; NTP, 1991). In developmental studies, effects on body weight and histopathological changes in the kidney were observed in fetuses of rats (Sprague-Dawley, Wistar and CD strains) exposed to oral doses or airborne concentrations of HCBD that also induced decreased body weight gain and/or renal effects in the dams (Schwetz *et al.*, 1977; Harleman and Seinen, 1979; Hardin *et al.*, 1981; Saillenfait *et al.*, 1989; NTP, 1990).

2.4.3.6 Neurological effects and effects on the immune system

Although data are limited, results of available short-term, subchronic and chronic studies in rodents do not indicate that neurological effects or effects on the immune system are critical endpoints associated with exposure to HCBD; that is, such effects were not observed at doses

lower than those that induced effects on the kidney (Kociba *et al.*, 1977a; Harleman and Seinen, 1979; Yang *et al.*, 1989; NTP, 1991). However, no studies on the effects of HCBD on the function of the immune system were identified.

2.4.3.7 Toxicokinetics and mechanism of action

The site-specific renal toxicity of HCBD is closely correlated with the accumulation of active metabolites in the pars recta of the proximal tubule. HCBD is initially conjugated with GSH in the liver to form sulphur conjugates, which are hydrolysed in the bile duct, intestine and kidney. These S-cysteine conjugates and their mercapturic acid derivatives (formed by N-acetylation) are concentrated in the kidney, where the pentachloro-sulphur conjugate is subsequently cleaved by renal β-lyase (which is localized in the pars recta) to reactive thiol metabolites, which may covalently bind to cellular macromolecules (causing cytotoxicity) and/or bind to DNA to induce mutation. (Note: Although metabolism of HCBD may be qualitatively similar in experimental animals and in humans, some very limited data indicate that the activity of β-lyase in the kidney of humans may be several-fold less than that in the kidney of rats; McCarthy *et al.*, 1992; Lock, 1994.) In addition, sulphoxidation of one of the mercapturic acid derivatives to electrophilic metabolites has been recently demonstrated in rats exposed to HCBD *in vivo* and in human liver microsomes (Birner *et al.*, 1995).

Although it is known that these electrophilic metabolites induce damage in renal tubular epithelial cells and mutations in *Salmonella* and bind to DNA, it has not been firmly established whether the initial step in kidney tumour formation is a result of genetic damage or epigenetic events (possibly in the mitochondria) (Stott *et al.*, 1981; Schrenk and Dekant, 1989; Dekant *et al.*, 1990; Henschler and Dekant, 1990). Unlike the mechanism of action associated with other halogenated hydrocarbons, accumulation of $\alpha_{2\mu}$-globulin and hyaline droplet formation are not involved in the formation of renal tumours induced by HCBD.

2.4.4 Humans

The limited identified studies in humans, which include a cross-sectional study on liver function and a survey of the frequency of chromosomal aberrations in exposed workers (German, 1986; Driscoll *et al.*, 1992), are inadequate to contribute meaningfully to evaluation of the toxicity of HCBD.

3.0 PROPOSED ASSESSMENT OF "TOXIC" UNDER CEPA 1999

3.1 CEPA 1999 64(a): Environment

The environmental risk assessment of a PSL substance is based on the procedures outlined in Environment Canada (1997a). Analysis of exposure pathways and subsequent identification of sensitive receptors are used to select environmental assessment endpoints (e.g., adverse reproductive effects on sensitive fish species in a community). For each endpoint, a conservative Estimated Exposure Value (EEV) is selected and an Estimated No-Effects Value (ENEV) is determined by dividing a Critical Toxicity Value (CTV) by an application factor. A conservative (or hyperconservative) quotient (EEV/ENEV) is calculated for each of the assessment endpoints in order to determine whether there is potential ecological risk in Canada. If these quotients are less than one, it can be concluded that the substance poses no significant risk to the environment, and the risk assessment is completed. If, however, the quotient is greater than one for a particular assessment endpoint, then the risk assessment for that endpoint proceeds to an analysis where more realistic assumptions are used and the probability and magnitude of effects are considered. This latter approach involves a more thorough consideration of sources of variability and uncertainty in the risk analysis.

There are special concerns about persistent and bioaccumulative substances. Persistent substances can remain bioavailable for long periods of time, increasing the probability and the duration of potential exposure. Even extremely low concentrations of persistent and bioaccumulative substances can have adverse effects on organisms that are continually exposed to them over long periods of time. Substances that are subject to long-range transport are of particular concern because cold regions, such as the Canadian Arctic, can act as a sink for such contaminants. Because of these concerns, environmental assessments of persistent and bioaccumulative substances are more conservative than those for other substances. Persistent and bioaccumulative substances may be determined to be toxic if they have the potential to harm the environment or its biological diversity, even if this is known to occur only within limited geographical areas within Canada.

3.1.1 Assessment endpoints

Current Canadian sources of HCBD are minor but potentially numerous. They include possible releases in landfill leachates, releases during refuse combustion and releases as a by-product in the production of other chlorinated chemicals. The most significant point source of HCBD in Canada appears to be the Cole Drain, which discharges into the St. Clair River at Sarnia, Ontario. Pelagic and benthic organisms are exposed to HCBD from this source. There is no indication that biota in Canadian marine systems are exposed to HCBD. Concentrations of HCBD in air and soil in Canada are generally low. The assessment endpoints for the environmental assessment of HCBD are normal growth and reproduction in populations of freshwater pelagic and benthic organisms in Canada.

3.1.2 Environmental risk characterization

3.1.2.1 Pelagic organisms

Concentrations of HCBD in St. Clair River water have declined considerably since the mid-1980s. The conservative EEV for pelagic organisms is 0.0027 µg/L, the highest reported concentration of HCBD in the St. Clair River in 1994.

The most sensitive freshwater species reported is the fathead minnow, with a 28-day LOEC of 13 µg/L, based on survival and growth. This value, 13 µg/L, is the conservative CTV for pelagic organisms. Dividing this CTV by a factor of 100 to account for uncertainty surrounding laboratory to field extrapolation and inter- and intraspecies differences in sensitivity gives an ENEV of 0.13 µg/L.

The conservative quotient is calculated by dividing the EEV of 0.0027 µg/L by the ENEV, as follows:

$$\text{Quotient} = \frac{\text{EEV}}{\text{ENEV}}$$

$$= \frac{0.0027\ \mu g/L}{0.13\ \mu g/L}$$

$$= 0.02$$

Because the conservative quotient is less than 1, this substance is unlikely to cause a harmful effect on populations of pelagic organisms in the ambient aquatic environment.

This quotient would be lower for freshwater invertebrates, since they appear to be somewhat less sensitive than fish to HCBD. The application factor of 100 used for deriving the ENEV is conservative, as the CTV was based on a 28-day LOEC, rather than a 96-hour LC_{50}.

The risk quotient for pelagic organisms is presented in Table 1.

3.1.2.2 Benthic organisms

The conservative EEV for benthic organisms is 243 µg/g dry weight, the highest reported concentration of HCBD in the top 5 cm of sediment in a 2-km stretch of the St. Clair River in an industrialized zone near Sarnia, Ontario, in 1994.

The CTV for benthic organisms is 20.8 µg/g dry weight, estimated using the Equilibrium Partitioning approach as presented in Section 2.4.1.2. Dividing this CTV by a factor of 100 to account for the uncertainty surrounding the extrapolation from laboratory to field conditions and interspecies and intraspecies variations in sensitivity gives an ENEV of 0.21 µg/g dry weight.

The conservative quotient is calculated by dividing the EEV of 243 µg/g by the ENEV, as follows:

$$\text{Quotient} = \frac{\text{EEV}}{\text{ENEV}}$$

$$= \frac{243\ \mu g/g}{0.21\ \mu g/g}$$

$$= 1157$$

Since the conservative quotient is more than 1, it is necessary to consider further the exposure of benthic biota to HCBD in the St. Clair River.

The cumulative density function for HCBD in St. Clair River sediments, at a depth of 0–5 cm, is shown in Figure 2. As stated in Section 2.3.2.4, the 99th-, 95th- and 90th-percentile values are 194, 60.9 and 18.7 µg/g dry weight, respectively, while the median is 0.9 µg/g dry weight.

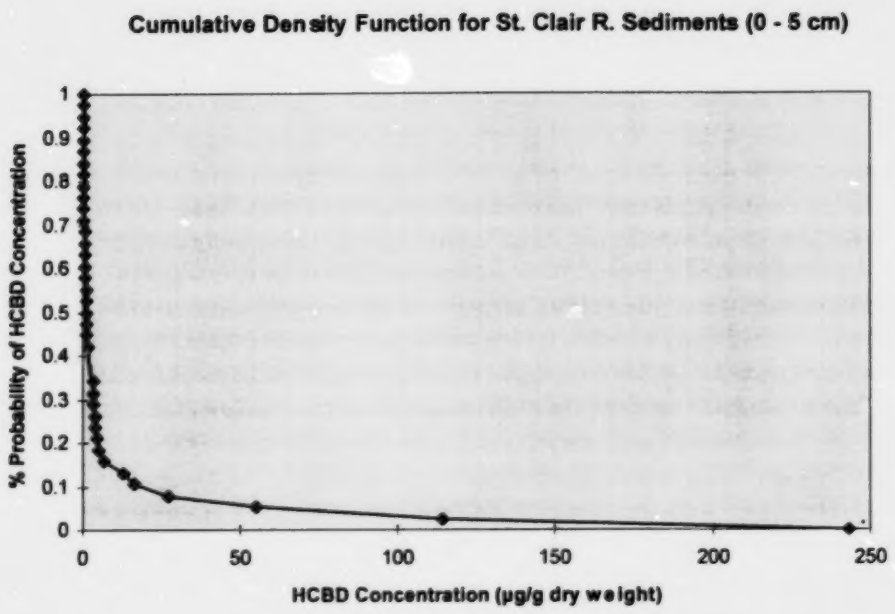


Figure 2: Cumulative density function for HCBD in St. Clair River sediments (0–5 cm)

Risk quotients for benthic organisms at various exposure levels in St. Clair River sediments are presented in Table 2. The ENEV in this table is the same as that used in the conservative risk assessment, 0.21 µg/g dry weight.

As indicated in Table 2, a quotient exceeding 1 occurs frequently in the sediments in the St. Clair River near Sarnia, Ontario. In fact, the concentration of HCBD in sediments in this area equalled or exceeded the ENEV of 0.21 µg/g dry weight at 29 of 39 sample stations. Benthic organisms in highly contaminated locations within this 2-km stretch of the St. Clair River could experience adverse effects because of their inability to move to less contaminated areas.

The sediment in this section of the St. Clair River contains a wide variety of organic and inorganic contaminants, including mercury, polychlorinated biphenyls, polychlorinated aromatic hydrocarbons, petroleum hydrocarbons and hexachlorobenzene, along with HCBD (Bedard and Petro, 1997). Whole-sediment toxicity tests were conducted on three species — the mayfly, *Hexagenia limbata* (21-day mortality and growth), the midge, *Chironomus tentans* (10-day mortality and growth), and the fathead minnow (21-day mortality) — using sediment samples taken from the most contaminated area. Significant correlations were found between lethality and HCBD concentration. HCBD bulk sediment concentrations explained 94% of the variation in midge mortality and 54% of the variation in mayfly mortality (Bedard and Petro, 1997). These results support the conclusion that benthic organisms in the most contaminated part of the St. Clair River can be harmed by HCBD in the sediments.

Because HCBD is persistent, with a half-life in air ranging from 60 days to 3 years and with a potential for long-range transport, as supported by measurements in Great Slave Lake sediments, and because the substance bioaccumulates, with a BCF ranging up to 19 000, a probabilistic risk assessment will not be performed. HCBD is still released to the environment in many sites, with concentrations in effluents up to 0.9 µg/L, compared with a pelagic ENEV of 0.13 µg/L.

3.1.2.3 Sources of uncertainty

There are several sources of uncertainty associated with the environmental assessment of HCBD. There were no acute or chronic toxicity studies using benthic organisms identified for HCBD. Effects on benthic organisms were therefore estimated using the Equilibrium Partitioning approach. This approach is based on the assumption that sediment interstitial water is the primary route of exposure of benthic organisms to HCBD, that continuous equilibrium exchange between sediment solids and interstitial water occurs, and that distribution of HCBD between these two phases can be estimated using the organic carbon/water partition coefficient of the substance and the organic carbon content of the sediment. Benthic organisms in highly contaminated areas of the St. Clair River at Sarnia, Ontario, may be adversely affected by HCBD, but the exact spatial extent of this area cannot be determined from existing data, because concentrations of the substance above the ENEV of 0.21 µg/g dry weight occurred at the sampling sites located farthest downstream. Concentrations of HCBD in sediments downstream from the source of contamination have been slowly declining since the mid-1980s.

3.1.2.4 Conclusion

The available information therefore indicates that HCBD poses little or no risk to pelagic aquatic organisms in Canada. HCBD poses a risk to benthic organisms in the most contaminated portions of the St. Clair River.

3.2 CEPA 1999 64(b): Environment upon which life depends

Worst-case calculations were made to determine if HCBD has the potential to contribute to depletion of stratospheric ozone, ground-level ozone formation or climate change. The Ozone Depletion Potential (ODP) was calculated to be 0.07, the POCP was estimated to be 0.01 and the GWP was calculated to be 0.037. These figures imply that HCBD is not likely to contribute significantly to ground-level ozone formation, but it does have the potential to contribute to depletion of stratospheric ozone and to climate change. Some substances currently subject to the Montreal Protocol have ODP values similar to the one calculated for HCBD, however, heres is general agreement that at these ODP values, substances should not be automatically subject to controls. Other criteria such as quantities emitted have to be part of the decision making. The concentration of HCBD in the Canadian atmosphere is low, estimates of its half-life in air based on photochemical degradation through reactions with hydroxyl radicals and ozone ranges from 60

days to three years.

Canadian sources of HCBD should not contribute significantly to depletion of stratospheric ozone or to climate change. HCBD is not produced or imported in Canada. Main Canadian sources are from combustion and as a by-product in the production of some chlorinated chemicals. Under the Montreal Protocol these sources are not subject to controls (incidentally produced substances).

According to the United States Toxic Release Inventory, 2 tonnes of HCBD were released to the environment in the United States in 1995; 75% of this total was to the air (Toxic Release Inventory, 1997). The load to the atmosphere, however, does not include all possible releases from every type of industrial facility (ATSDR, 1994). HCBD is also on the OECD high production volume list, which means it is produced in excess of 10 000 tonnes per year in at least one OECD country (SIDS Manual, 1994). Limited information on quantities, concentrations or conditions of foreign sources of HCBD prevent us from reaching an overall conclusion on the danger to the environment on which life depends.

3.3 CEPA 1999 64(c): Human health

3.3.1 Estimated population exposure

Available data on levels of HCBD in environmental media in Canada upon which estimates of population exposure may be based are quite limited. Point estimates of average daily HCBD intake (on a body weight basis), based on the data on levels of HCBD in ambient air, drinking water and food summarized in Section 2.3.2 and reference values for body weight, inhalation volume and amounts of drinking water and food consumed daily, are presented for five age groups in Table 3.[1] Based on these estimates, intake may range from 0.01 to 0.2 µg/kg-bw per day. However, it should be noted that these estimates are based on very few samples of only a small number of foodstuffs in early studies in other countries or primarily on limits of detection (or one-half the detection limit for air) in monitoring surveys in which HCBD was only rarely detected in other media. They are presented primarily, therefore, for the purpose of identifying the potential relative contribution of these media to overall population exposure.

If estimates were based on the limit of detection for food and beverages in the limited pilot multimedia study in Toronto in which HCBD was not detected, estimated intakes would be similar to values at the upper end of the range presented in Table 3.

---

[1] The exposure assessment for HCBD was completed prior to the characterization of intake values for six age groups, which is the approach that will be adopted for the remainder of the substances on the second Priority Substances List (PSL2). However, to the extent possible, recent information relevant to the development of intakes for six age groups for PSL2 substances has been taken into account as described in Appendix C of the supporting documentation for the health-related sections.

Based on the values derived by either approach, food (or food and beverages) is likely the principal source of exposure, although ambient air may also contribute significant amounts in some areas; drinking water contributes negligibly to overall intake of HCBD. This is consistent with apportionment predicted on the basis of physical/chemical properties or fugacity modelling, although the latter was not helpful in further refinement of estimation of exposure because of a lack of quantitative data on emissions of HCBD into the Canadian environment.

In order to examine the distribution of population exposure to HCBD in Canada, probabilistic estimates were also derived for each of the five age groups, based on information on the distribution of body weights and inhalation volumes, as well as data from the national survey of concentrations of HCBD in ambient air. Data were inadequate to derive probabilistic exposure estimates for other media (i.e., drinking water or food). Estimates of mean, median and 95th-percentile intakes are included in Table 4, along with the point estimates derived in Table 3 for comparison. For example, the 95th-percentile estimates of intake from air range from 0.03 to 0.09 µg/kg-bw per day, compared with point estimates of 0.01–0.05 µg/kg-bw per day.

3.3.2 Hazard characterization

Because of the inadequacy of data in humans, hazard characterization and dose–response analysis for HCBD are based on studies in experimental animals.

In acute, short-term, subchronic and chronic studies in rats and mice exposed to HCBD via ingestion or inhalation, effects in the pars recta of the proximal tubules of the kidneys (including increased organ weights and biochemical and histopathological evidence of degeneration) consistently occur at the lowest dose or concentration that caused effects (Kociba *et al.*, 1971, 1977a; Schwetz *et al.*, 1977; Harleman and Seinen, 1979; Stott *et al.*, 1981; Yang *et al.*, 1989; NTP, 1991; Jonker *et al.*, 1993; Birner *et al.*, 1995).

There was also an increased incidence of renal tubular tumours in male and female Sprague-Dawley rats administered the highest dose of HCBD in the diet for two years; nephrotoxicity in the form of hyperplasia and adenomatous proliferation in the renal tubular epithelium was also observed at this as well as a lower dose (Kociba *et al.*, 1977a). Unlike the mechanism of action associated with other halogenated hydrocarbons, accumulation of $\alpha_{2\mu}$-globulin and hyaline droplet formation are not involved in the formation of renal tumours induced by HCBD.

The weight of available evidence indicates that HCBD is genotoxic in the presence of appropriate metabolic activation systems (Reichert *et al.*, 1984; Vamvakas *et al.*, 1988). This is consistent with the increased incidence of renal tumours observed in rats *in vivo*, binding of HCBD metabolites to kidney mitochondrial DNA in mice and small amounts of DNA alkylation in the kidney of rats (Stott *et al.*, 1981; Schrenk and Dekant, 1989).

Both genotoxic and non-genotoxic steps may be involved in the induction of tumours by HCBD, although the critical rate-limiting step has not been identified. However, based on observations in the single adequate carcinogenesis bioassay, tumours occur only at doses greater than those that induce non-neoplastic effects in the kidney. These degenerative effects and resulting regeneration are likely requisite in the induction of tumours and are considered, therefore, to be the critical endpoint. The renal toxicity of HCBD is closely correlated with the site specificity of accumulation of active metabolites, and there is some (albeit limited) evidence that extent of activation may be less in humans than in rats (e.g., cleavage of the cysteine conjugate by renal β-lyase) (Lock, 1994).

Based on limited data, reproductive and developmental effects and neurotoxicity are not considered to be critical endpoints for HCBD, since effects have been observed only at doses greater than those associated with renal toxicity. Data on effects of HCBD on immunological function have not been identified.

3.3.3 Dose–response analyses

Since non-neoplastic renal effects observed in experimental animals are considered critical and since available data are sufficient, a Tolerable Intake (TI) is derived on the basis of a benchmark dose (BMD) divided by an uncertainty factor. This value is compared with that which might be based on a No-Observed-(Adverse)-Effect-Level (NO[A]EL) for this endpoint, which draws on data from additional studies.

In the available short-term, subchronic and chronic studies, the kidney has consistently been observed to be the most sensitive target organ, with similar effect levels noted in the critical studies (Table 5). In the only identified long-term study in which animals were exposed via ingestion (Kociba *et al.*, 1977a), an increased incidence of renal tubular hyperplasia/proliferation and an increase in levels of renal coproporphyrin were observed in Sprague-Dawley rats administered 2.0 mg HCBD/kg-bw per day (considered to be the LO[A]EL) or more; renal tubular neoplasms were observed at the highest dose of 20 mg/kg-bw per day. The NOEL was considered to be 0.2 mg/kg-bw per day. Similarly, the LOEL and NOEL for renal toxicity (renal tubular dilation and hypertrophy with foci of renal tubular epithelial degeneration and regeneration) in a subchronic study in the same strain of rats (i.e., Sprague-Dawley) were also 2.0 and 0.2 mg/kg-bw per day, respectively (Schwetz *et al.*, 1977). Renal tubular regeneration (of a severity greater than would be expected, based on comparison with data for the next dose group) also occurred in 1 of 10 mice at the lowest dose tested in a subchronic study in $B6C3F_1$ mice, 0.2 mg/kg-bw per day (Yang *et al.*, 1989; NTP, 1991), which is considered to be the LOEL. In two of these studies (Harleman and Seinen, 1979; Jonker *et al.*, 1993), decreases in body weight (generally associated with reduced food consumption) were also observed at the LOAEL for renal toxicity.

Sufficient information to permit modelling of the dose–response curve for development of a BMD for renal toxicity was presented in few of these studies. The endpoint that is most amenable to derivation of a BMD is the renal tubular regeneration observed in the 13-week study in B6C3F$_1$ mice (Yang *et al.*, 1989; NTP, 1991), in which the incidence of this lesion is presented for each dose group. Using the THRESH program, which fits a polynomial model to the data, the BMD_{05} (the dose associated with a 5% increase in the incidence of renal tubular regeneration) for female mice (which were observed to be more sensitive than males) was 160 µg/kg-bw per day ($\chi^2 = 0$, df = 0, p = 1.0). The 95% lower confidence limit on this value ($BMDL_{05}$) is 34 µg/kg-bw per day. BMDs calculated for other endpoints in the available subchronic and chronic studies, although based on very limited data in some cases, were greater than those for renal tubular regeneration in female mice presented here.

A TI has been developed on the basis of the $BMDL_{05}$ for renal tubular regeneration in mice as follows:

$$TI = \frac{34 \text{ µg/kg-bw per day}}{100}$$

$$= 0.34 \text{ µg/kg-bw per day}$$

where:

- 34 µg/kg-bw per day is the 95% lower confidence limit of the dose estimated to be associated with a 5% increase in renal tubular regeneration in mice administered HCBD for 13 weeks (Yang *et al.*, 1989; NTP, 1991), and
- 100^2 is the uncertainty factor (×10 for interspecies variation and ×10 for intraspecies variation; the default values are applied since limited available data on pharmacokinetics and pharmacodynamics in the experimental species and humans are considered insufficient to derive more appropriate values, although the 10-fold factor for interspecies variation is slightly less than a value that would be developed on the basis of the surface area to body weight correction for this species).

This TI is protective, based on consideration of the NOEL for renal toxicity of 0.2 mg/kg-bw per day observed in the chronic study in rats (Kociba *et al.*, 1977a) and supported by the results of the subchronic studies in rats and mice in which a NOEL and LOEL, respectively, of the same value were observed (Schwetz *et al.*, 1977; Yang *et al.*, 1989; NTP, 1991). Although the variation between doses in the study by Kociba *et al.* (1977a) was large (i.e., 10-fold), it was less in the investigation in mice (i.e., 3-fold). Based on application of the same uncertainty factor

[2] An additional factor for use of the LOEL was not incorporated, since the renal lesion was observed in only 1 of 10 females in the lowest dose group (not statistically significant); inadequate data were available to determine whether this response may have been a function of, for example, increased food consumption in this single animal.

applied in the derivation of the TI above (i.e., 100) to the NOEL of 0.2 mg/kg-bw per day, the resulting value is greater than 0.34 µg/kg-bw per day (i.e., 2 µg/kg-bw per day).

Available data on the effects associated with inhalation of HCBD are much more limited than those for ingestion. The only relevant studies identified include a short-term study in which renal toxicity was observed in rats exposed to concentrations of 25 ppm (267 mg/m^3) HCBD and above for up to 15 days (NOEL = 5 ppm or 53 mg/m^3) (Gage, 1970) and a developmental study in which reductions in maternal weight gain were observed in rats exposed to 5 ppm (53 mg/m^3) and above (Saillenfait *et al.*, 1989). (Interpretation of this latter observation is complicated by the absence of an exposure–response relationship and the lack of presentation of data on food consumption.) Both of these studies are considered to be inadequate to serve as a basis for derivation of a Tolerable Concentration (TC) in air. If derived on the basis of the limited existing data, however, such values would, in any case, be greater than that developed above for ingestion. Derivation of a TC through conversion of the TI presented above is not considered appropriate in view of the absence of data on the kinetics of inhaled HCBD (i.e., based on available information, it is not possible to determine if the inhaled substance behaves in a manner similar to that following ingestion). However, it is noteworthy that renal toxicity was the critical effect in the limited short-term inhalation study in rats.

3.3.4 Human health risk characterization

Based on the point estimates of exposure for the various age groups derived from limited available monitoring data, highly uncertain average total daily intakes of HCBD from air, food and drinking water range from 0.01 to 0.2 µg/kg-bw per day. "Reasonable worst-case" estimates also fall within this range. These estimates are based, for the likely principal medium of exposure, primarily on monitoring data for a small number of foodstuffs for which there is considerable uncertainty about the extent of representation of current exposure of the Canadian public. Some of these data were obtained from industrial areas of other countries at a time when releases of HCBD into the ambient environment were likely much greater than current releases. This is offset to some extent by assumed zero exposure from foodstuffs for which data on concentrations were not available. Moreover, although levels of HCBD in ambient air in Canada have been well characterized in a national survey, it should be noted that estimated intake in this medium is based on half detection limits in the vast majority of samples (>98%) in which HCBD was not detected. In view of these limitations, it is reassuring that the maximum value for estimated average total daily intake and reasonable worst-case estimates (i.e., estimates based on the pilot multimedia study in Toronto in which HCBD was not detected in any medium) of 0.2 µg/kg-bw per day, although also uncertain, is still less than the TI of 0.34 µg/kg-bw per day calculated from the 95% lower confidence limit of the BMD for effects in the kidney in subchronically exposed mice. It should be further noted that this TI is considered conservative, based on a value that might be derived on the basis of a NOEL for renal toxicity in rats exposed to HCBD for two years.

Therefore, on the basis of comparison of estimates of exposure and the TI (i.e., intakes to which it is believed that a person may be exposed daily over a lifetime without deleterious effects), it has been concluded that HCBD is not present in the environment in quantities or under conditions that may constitute a danger in Canada to human life or health.

3.3.5 Uncertainties and degree of confidence in human health risk characterization

There is a high degree of uncertainty inherent in the estimates of intake of HCBD in food, the likely principal medium of exposure, because of the limited number of foodstuffs for which monitoring data are available and the fact that those data that are available were often acquired in early surveys in other countries. There is also considerable uncertainty in the reasonable worst-case estimates for food due to the lack of determination of analytical recovery in the multimedia study.

Although confidence in the estimates of intake in air is greater, since levels of HCBD in ambient air in Canada have been well characterized in a national survey, a degree of uncertainty is introduced by the assumption of half detection limits in the vast majority of samples in which HCBD was not detected. This degree of uncertainty has been characterized quantitatively by calculating intakes also on the basis of the assumption of zero or detection limit for measurements below the detection limit in the national survey. Maximum values for estimated average intakes from air would be approximately one-third of those presented based on an assumption of zero for non-detectable concentrations and twice those presented based on the assumption of detection limit for these samples.

However, there is a high degree of certainty that drinking water contributes only negligible amounts of HCBD to overall exposure, based on the number of large, sensitive investigations.

The only route for which probabilistic estimates of exposure could be derived was inhalation via ambient air. Based on these estimates, intake of HCBD by 95% of the age group with the greatest intake per unit of body weight (i.e., 0.5–4 year olds) is about twice the (uncertain) point estimate for intake via inhalation (i.e., 0.09 µg/kg-bw per day versus 0.04–0.05 µg/kg-bw per day).

In addition, fugacity modelling was not helpful in refinement of estimation of exposure due to the lack of quantitative data on emissions of HCBD into the Canadian environment.

The overall degree of confidence in the population exposure estimates is, therefore, low, primarily as a result of the paucity of current, representative monitoring data for the likely principal medium of exposure of the general population in Canada (food).

The degree of confidence in the database on toxicity that serves as the basis for development of the TI is moderate to high. Although epidemiological data in humans are

inadequate, there is consistent evidence from a wide range of acute, short-term, subchronic and chronic studies in rats and mice that critical effects are those that occur in the pars recta of the renal proximal tubules, although data on reproductive effects are somewhat limited, and information on effects on immunological function has not been identified. Moreover, the range of lowest-effect levels at which degenerative renal changes have been observed in long-term studies (subchronic and chronic) is small, and available data are sufficient to develop a BMD and associated lower 95% confidence interval for such effects. Although there is some uncertainty about the mode of induction of tumours by HCBD observed in a single study, there is reasonable assurance that tumours occur only in the presence of degenerative renal changes.

3.4 Proposed conclusions

CEPA 1999 64(a): Based on available data, it has been concluded that HCBD is entering the environment in a quantity or concentration or under conditions that have an immediate or long-term harmful effect on the environment or its biological diversity.

CEPA 1999 64(b): Based on available data, it has been concluded that HCBD is not entering the environment, in Canada, in a quantity or concentration or under conditions that constitute a danger to the environment on which life depends.

CEPA 1999 64(c): Based on available data, it has been concluded that HCBD does not occur in the environment in quantities or concentrations that constitute or may constitute a danger in Canada to human life or health.

Proposed overall conclusion: Based on critical assessment of relevant information, it is proposed that HCBD be considered as "toxic" as defined in Section 64 of CEPA 1999.

3.5 Considerations for follow-up (further action)

Pursuant to subsection 77(4), because HCBD is proposed to be toxic under the Act and meets the criteria for persistence and bioaccumulation in accordance with the *Persistence and Bioaccumulation Regulations*, is present in the environment primarily as a result of human activity, and is not a naturally-occurring radionuclide or a naturally-occurring inorganic substance, implementation of virtual elimination under subsection 65(3) of HCBD is being proposed.

It is recommended that sources of HCBD as a by-product in the production of other chlorinated chemicals, such as vinyl chloride, allyl chloride and epichlorohydrin be identified and measures to reduce these releases be investigated.

HCBD releases during refuse combustion were identified. Preliminary information indicates that sources of HCBD from combustion are similar to those of dioxins, furans and hexachlorobenzene. It is recommended that measures to reduce emission of HCBD from combustion sources complement initiatives currently underway to address dioxins, furans and hexachlorobenzene.

Since HCBD is persistent, bioaccumulative, likely to cause effects to aquatic species at low levels of exposure and is not currently used in commerce in Canada, options to prevent its reintroduction into the Canadian market should be explored.

One potential source of HCBD in Canada identified in the current assessment is transboundary movement from foreign sources. It is recommended, therefore, that the significance of this source be considered in the context of international programs addressing long-range transport of transboundary pollutants.

Table 1: Risk quotient for pelagic organisms

Parameter	**Value**
EEV	0.0027 µg/L
CTV	13 µg/L
Application factor	100
ENEV	0.13 µg/L
Quotient (EEV/ENEV)	0.02

Table 2: Summary of risk quotients for freshwater benthic organisms

EEV (µg/g dry weight)	Descriptor	CTV (µg/g)	Application factor	ENEV (µg/g)	Quotient (EEV/ENEV)
243	Maximum reported concentration, 1994	20.8	100	0.21	1157
194	99th percentile, 1994	20.8	100	0.21	924
60.9	95th percentile, 1994	20.8	100	0.21	290
18.7	90th percentile, 1994	20.8	100	0.21	89
0.9	Median, 1994	20.8	100	0.21	4.3

Table 3: Estimated exposure of the general population to HCBD

Medium	Estimated intake (µg/kg-bw per day)				
	0–0.5 years[1]	0.5–4 years[2]	5–11 years[3]	12–19 years[4]	20–70 years[5]
Air[6]	<0.02–0.02	0.04–0.05	0.03–0.04	0.01–0.02	0.01–0.02
Drinking water[7]	<0.0001	<0.000 06	<0.000 03	<0.000 02	<0.000 02
Food[8]	0.03–0.07[9]	0.004–0.1	0.001–0.05	0.0009–0.03	0.001–0.03
Total	<0.05–0.09	0.04–0.2	0.03–0.09	0.01–0.05	0.01–0.05

1 Assumed to weigh 7 kg, to drink 0.75 L of water per day (Health Canada, 1994) and to breathe 2.1 m^3 air per day.
2 Assumed to weigh 13 kg, to drink 0.8 L of water per day (Health Canada, 1994) and to breathe 9.3 m^3 air per day.
3 Assumed to weigh 27 kg, to drink 0.9 L of water per day (Health Canada, 1994) and to breathe 14.5 m^3 air per day.
4 Assumed to weigh 57 kg, to drink 1.3 L of water per day (Health Canada, 1994) and to breathe 15.8 m^3 air per day.
5 Assumed to weigh 70 kg, to drink 1.5 L of water per day (Health Canada, 1994) and to breathe 15.8 m^3 air per day.
6 Based on the range of mean concentrations of HCBD in ambient air in 46 locations across Canada of 0.05–0.0 µg/m^3 (Dann, 1997). HCBD was not detected in 98% of these ambient air samples. A concentration of 0.0 µg/m^3 (which is one-half the limit of detection of 0.1 µg/m^3) was assumed for the samples in which HCBD was not detected. As no adequate data were identified on levels of HCBD in indoor air, it is assumed that the concentrations of HCBD in indoor and outdoor air are similar.
7 Based on the assumption that HCBD is present at concentrations less than the detection limit of 0.00 µg/L reported in the largest of the available surveys of drinking water supplies in Canada (Graham, 1993).
8 Based on concentrations of HCBD reported for various foodstuffs in the United States (Yip, 1976), the United Kingdom (McConnell *et al.* 1975) and Germany (Kotzias *et al*, 1975), limited data on levels in fish caught in Canada (Fox *et al.*, 1983; Oliver and Niimi, 1983), the United States (Oliver and Nicol, 1982; Clark *et al.*, 1984; Malins *et al*, 1985) and the Netherlands (Goldbach *et al.*, 1976) and average daily food consumption patterns per age group (Health Canada, 1994). In all other food types, minimum concentrations are assumed to be zero. In 8 of the 14 food types on which estimates are based, minimum values are considered to be zero (whole milk, butter, eggs, fish [marine], cabbage, beans, cucumbers and margarine); in the remainder (evaporated milk, fish [freshwater], tomatoes, grapes, vegetable oil and alcoholic drinks), minimum values were the lowest measured or the single concentration reported. In 10 of the 14 food types on which estimates are based, maximum values are either the highest reported concentration (for 5 of the food types — whole milk, butter, eggs, fish [marine] and margarine) or the single concentration reported (for 5 of the food types — evaporated milk, tomatoes, grapes, vegetable oil and alcoholic drinks). A maximum concentration equivalent to the limit of detection (µg/kg) was assumed for 3 food types (vegetables) based on the analyses of Yip (1976). A maximum concentration (10 µg/kg) in freshwater fish obtained from non-source-dominated areas of North America was assumed (Clark *et al.*, 1984). Data from freshwater fish samples collected in source-dominated areas in countries other than Canada were not considered relevant.
9 Based on the assumption that infants were exclusively fed prepared foodstuff. If it is assumed that infants are exclusively breast-fed and consume an average of 0.75 L/day (Health Canada, 1994) and that HCBD is present in breast milk at the detection limit of µg/L reported for Canadian women (Mes *et al.*, 1986), the average daily intake by ingestion is <0.13 µg/kg-bw per day.

Note: Insufficient data were available with which to estimate intake from soil.

Table 4: Point versus probabilistic estimates of exposure to HCBD via inhalation

Approach	Parameter estimated	Estimated intake by inhalation (µg/kg-bw per day)				
		0–0.5 years[2]	0.5–4 years[3]	5–11 years[4]	12–19 years[5]	20–70 years[6]
Point estimate	Average daily intake	0.02	0.04–0.05	0.03–0.04	0.01–0.02	0.01–0.02
Probabilistic	Median intake	0.01	0.03	0.03	0.01	0.01
Probabilistic	Mean intake	0.02	0.04	0.03	0.02	0.01
Probabilistic	95th-percentile intake	0.04	0.09	0.06	0.03	0.03

[1] Point estimates are based on the range of mean concentrations of HCBD in ambient air in 46 locations across Canada of 0.05–0.07 µg/m^3 (Dann, 1997). HCBD was not detected in 98% of the 9231 ambient air samples. A concentration of 0.05 µg/m^3 (which is one-half the limit of detection of 0.1µg/m^3) was assumed for the samples in which HCBD was not detected. Probabilistic estimates are based on Monte Carlo simulations with random sampling of HCBD concentrations from the distribution of reported concentrations in 9231 samples. All HCBD concentrations between 0 and 0.1 µg/m^3 (i.e., the limit of detection) are assumed to occur with the same probability (i.e., a uniform distribution of concentrations below the limit of detection is assumed). HCBD concentrations greater than 0.1µg/m^3 are sampled at the relative frequencies with which they occur among the 9231 samples. As no adequate data were identified on levels of HCBD in indoor air, it is assumed that the concentrations of HCBD in indoor and outdoor air are similar.

[2] Assumed to weigh 7 kg, to drink 0.75 L of water per day (Health Canada, 1994) and to breathe 2.1 m^3 air per day.

[3] Assumed to weigh 13 kg, to drink 0.8 L of water per day (Health Canada, 1994) and to breathe 9.3 m^3 air per day.

[4] Assumed to weigh 27 kg, to drink 0.9 L of water per day (Health Canada, 1994) and to breathe 14.5 m^3 air per day.

[5] Assumed to weigh 57 kg, to drink 1.3 L of water per day (Health Canada, 1994) and to breathe 15.8 m^3 air per day.

[6] Assumed to weigh 70 kg, to drink 1.5 L of water per day (Health Canada, 1994) and to breathe 15.8 m^3 air per day.

Table 5: Critical studies and effect levels for renal toxicity in experimental animals exposed to HCBD via ingestion

Species	Protocol	Effects at LO(A)EL	Effect levels	Comments	Reference
Wistar rats (5 males and 5 females per group)	Rats were exposed to doses of 0, 1.25, 5 or 20 mg/kg-bw per day in the diet for 4 weeks	Decreased body weight and food consumption; increased relative kidney weight; decreased relative weight of adrenals; effects on urinary and biochemical parameters; histopathological effects in kidney	NOAEL (females) = 1.25 mg/kg-bw per day LOAEL (females) = 5 mg/kg-bw per day NOAEL (males) = 1.25 mg/kg-bw per day LOEL (males) = 5 mg/kg-bw per day	Small number of animals per group	Jonker *et al.*, 1993
Wistar rats (10 males and 10 females per group)	Rats were exposed to doses of 0, 0.4, 1.0, 2.5, 6.3 or 15.6 mg/kg-bw per day by gavage for 13 weeks	Effects on urinary parameters; histopathological effects in kidney	NOEL (females) = 1.0 mg/kg-bw per day LOAEL (females) = 2.5 mg/kg-bw per day NOEL (males) = 2.5 mg/kg-bw per day LOAEL (males) = 6.3 mg/kg-bw per day	Small number of animals per group; large number of dose groups with good spacing between dose levels	Harleman and Seinen, 1979
Sprague-Dawley rats (10–12 males and 20–24 females per group; 17 male and 34 female controls)	Rats were exposed to doses of 0, 0.2, 2.0 or 20 mg/kg-bw per day in the diet for about 5 months	Gross and histopathological changes in kidney	NOEL = 0.2 mg/kg-bw per day LOEL = 2.0 mg/kg-bw per day	Small number of animals per group	Schwetz *et al.*, 1977
Sprague-Dawley rats (39–49 males and 40 females per group; 90 male and 90 female controls)	Rats were exposed to doses of 0, 0.2, 2.0 or 20 mg/kg-bw per day in the diet for 2 years	Effects on urinary biochemical parameters; histopathological effects in kidney	NOEL = 0.2 mg/kg-bw per day LO(A)EL = 2.0 mg/kg-bw per day	Good study protocol, except for dose spacing; description of non-neoplastic effects incomplete	Kociba *et al.*, 1977a
B6C3F$_1$ mice (10 males and 10 females per group)	Mice were exposed to doses of 0, 0.1, 0.4, 1.5, 4.9 or 16.8 (males) or 0, 0.2, 0.5, 1.8, 4.5 or 19.2 (females) mg/kg-bw per day in the diet for 13 weeks	Histopathological effects in kidney	LOEL (females) = 0.2 mg/kg-bw per day NOAEL (males) = 1.5 mg/kg-bw per day	Small number of animals per group; large number of exposure groups with good dose spacing	Yang *et al.*, 1989; NTP, 1991

4.0 REFERENCES

Alberta Environmental Protection. 1996. Data summary prepared for the Priority Substances Section, Environmental Protection Services, Municipal Water and Wastewater Branch, Air and Water Approval Division, Edmonton, Alberta.

Anders, M.W. and I. Jakobson. 1985. Biotransformation of halogenated solvents. International Conference on Organic Solvent Toxicity, Stockholm, Sweden, October 15–17, 1984. Scand. J. Work Environ. Health 11(Suppl. 1): 23–32.

ATSDR (Agency for Toxic Substances and Disease Registry). 1994. Toxicological profile for hexachlorobutadiene. Public Health Service, U.S. Department of Health and Human Services (Publication No. TP-93/08).

Bedard, D. and S. Petro. 1997. Laboratory sediment bioassay report on Upper St. Clair River sediments in the vicinity of industrial point sources — 1994 & 1995. St. Clair River Remedial Action Plan, Toronto, Ontario. 76 pp.

Benoit, D.A., F.A. Puglisi and D.L. Olson. 1982. A fathead minnow (*Pimephales promelas*) early life stage toxicity test method evaluation and exposure to 4 organic chemicals. Environ. Pollut. Ser. A, Ecol. Biol. 28: 189–198.

Birner, G., M. Werner, M.M. Ott and W. Dekant. 1995. Sex differences in hexachlorobutadiene biotransformation and nephrotoxicity. Toxicol. Appl. Pharmacol. 132: 203–212.

Brown, S.L., F.Y. Chan, J.L. Jones, D.H. Liu, K.E. McCaleb, T. Mill, K.N. Sapios and D.E. Schendel. 1975. Research program on hazard priority ranking of manufactured chemicals (chemicals 1–20). Stanford Research Institute, Menlo Park, California (NSF/RA/E-75/190A.-PB263161).

Bunce, N. 1996. Atmospheric properties of substances on the Priority Substances List #2 (PSL2). Report to Environment Canada. University of Guelph, Guelph, Ontario. 13 pp.

Chan, C.H. 1993. St. Clair River head and mouth water quality monitoring, 1987–89. Water Pollut. Res. J. Can. 28(2): 451–471.

Chiou, C.T. 1985. Partition coefficients of organic compounds in lipid–water systems and correlations with fish bioconcentration factors. Environ. Sci. Technol. 19(1): 57–62.

Choudhary, G. 1995. Human health perspectives on environmental exposure to hexachlorobutadiene: a review. Environ. Carcinogen. Ecotoxicol. Rev. C13(2): 179–203.

Clark, J.R., D. DeVault, R.J. Bowden and J.A. Weishaar. 1984. Contaminant analysis of fillets from Great Lakes coho salmon, 1980. J. Great Lakes Res. 10(1): 38–47.

Class, T. and K. Ballschmiter. 1987. Global baseline pollution studies. X. Atmospheric halocarbons: global budget estimations for tetrachloroethene, 1,2-dichloroethane, 1,1,1,2-tetrachloroethane, hexachloroethane, hexachlorobutadiene. Estimation of the hydroxyl radical concentrations in the troposphere of the Northern and Southern Hemisphere. Fresenius Z. Anal. Chem. 327(2): 198–204.

Dann, T. 1997. Summary of hexachlorobutadiene concentrations at Canadian sites. Air Toxics Section, Environmental Technology Service, Environmental Protection Service, Environment Canada.

Dekant, W., S. Vamvakas, K. Berthold, S. Schmidt, D. Wild and D. Henschler. 1986. Bacterial β-lyase mediated cleavage and mutagenicity of cysteine conjugates derived from the nephrocarcinogenic alkenes, trichloroethylene, tetrachloroethylene, and hexachlorobutadiene. Chem. Biol. Interact. 60: 31–45.

Dekant, W., S. Vamvakas, M. Koob, A. Kochlin, W. Kanhai, D. Muller and D. Henschler. 1990. A mechanism of haloalkene-induced renal carcinogenesis. Environ. Health Perspect. 88: 107–110.

De Meester, C., M. Mercier and F. Poncelet. 1980. Mutagenic activity of butadiene, hexachlorobutadiene, and isoprene. Ind. Environ. Xenobiotics, Proc. Int. Conf. pp. 195–203 [cited in ATSDR, 1994].

Di Toro, D.M., C.S. Zarba, D.J. Hansen, W.J. Berry, R.C. Swartz, C.E. Cowan, S.P. Pavlou, H.E. Allen, N.A. Thomas and P.R. Paquin. 1991. Technical basis for establishing sediment quality criteria for nonionic organic chemicals using equilibrium partitioning. Environ. Toxicol. Chem. 10: 1541–1583.

DMER (Don Mackey Environmental Research) and AEL (Angus Environmental Limited). 1996. Pathways analysis of hexachlorobutadiene for the second Priority Substances List using fugacity modelling. Report prepared for the Chemicals Evaluation Division, Commercial Chemicals Evaluation Branch, Environment Canada, by DMER, Peterborough, Ontario, and AEL, Don Mills, Ontario, March 1996.

Dow Chemical Co. 1978. The acute fish toxicity of hexachlorobutadiene and hexachloroethane to the sheepshead minnow (*Cyprinodon variegatus*). The Dow Chemical Company, Midland, Michigan (TWC 103).

Driscoll, T.R., H.H. Hamdan, G. Wang, P.F.A. Wright and N.H. Stacey. 1992. Concentrations of individual serum or plasma bile acids in workers exposed to chlorinated aliphatic hydrocarbons. Br. J. Ind. Med. 49: 700–705.

Elwell, M.R. 1993. Personal communication with B. Mintz, U.S. Environmental Protection Agency. Letter dated April 21, 1993, from M.R. Elwell, National Institute of Environmental Health Sciences, Research Triangle Park, North Carolina.

Environment Canada. 1979. Substances in the list of priority chemicals — 1979. Commercial Chemicals Branch, Ottawa, Ontario (CCB-IN-3-80).

Environment Canada. 1983. Hexachlorobutadiene. *In:* Guidelines for surface water quality. Volume 2. Organic chemical substances. Water Quality Branch, Inland Waters Directorate, Ottawa, Ontario.

Environment Canada. 1997a. Environmental assessments of Priority Substances under the *Canadian Environmental Protection Act*. Guidance manual version 1.0 — March 1997. Chemicals Evaluation Division, Commercial Chemicals Evaluation Branch, Ottawa, Ontario (EPS 2/CC/3E).

Environment Canada. 1997b. CEPA Section 16 Notice to Industry respecting the second Priority Substances List and di(2-ethylhexyl)phthalate. Use Patterns Section, Commercial Chemicals Evaluation Branch, Hull, Quebec.

Environment Canada. 1997c. Results of the CEPA Section 16 Notice to Industry respecting the second Priority Substances List and di(2-ethylhexyl)phthalate. Use Patterns Section, Commercial Chemicals Evaluation Branch, Hull, Quebec.

Environment Canada. 1999. *Canadian Environmental Protection Act* — Priority Substances List supporting document for the environmental assessment of hexachlorobutadiene. Commercial Chemicals Evaluation Branch, Hull, Quebec.

Environment Canada, United States Environmental Protection Agency, Ontario Ministry of the Environment and New York Department of Environmental Conservation. 1995. Joint evaluation of upstream/downstream Niagara River monitoring data for the period April 1993 to March 1994. Report prepared by the River Monitoring Committee, Niagara River Data Interpretation Group.

Environment Ontario. 1987. Ottawa (Lemieux Island) Water Treatment Plant. Annual report 1987.

Farara, D.G. and A.J. Burt. 1997. Assessment of Upper St. Clair River sediments and benthic macroinvertebrate communities — 1994. Report prepared by Beak Consultants Ltd. for the Ontario Ministry of Environment and Energy, Toronto, Ontario.

Fox, M.E., J.H. Carey and B.G. Oliver. 1983. Compartmental distribution of organochlorine contaminants in the Niagara River and the western basin of Lake Ontario. J. Great Lakes Res. 9: 287–294.

Fragiadakis, A., W. Klein, F. Korte, P.N. Moza, I. Scheunert, D. Vockel and U. Weiss. 1979. [Behaviour of organohalogen compounds in the plant–soil systems.] Spez. Ber. Kernforschungsanlage Juelich 45: 144–155 (in German).

Gage, J.C. 1970. The subacute inhalation toxicity of 109 individual chemicals. Br. J. Ind. Med. 27: 1–8.

Galloway, S.M., M.J. Armstrong, C. Reuben, S. Colman, B. Brown, C. Cannon, A.D. Bloom, F. Nakamura, M. Ahmed, S. Duk, J. Rimpo, B.H. Margolin, M.A. Resnick, B. Anderson and E. Zeiger. 1987. Chromosome aberrations and sister chromatid exchanges in Chinese hamster ovary cells: Evaluation of 108 chemicals. Environ. Mol. Mutagen. 10 (Suppl. 10): 1–175.

Geiger, D.L., C.E. Northcott, D.J. Call and L.T. Brooke. 1985. Acute toxicities of organic chemicals to fathead minnows (*Pimephales promelas*). Volume II. Center for Lake Superior Environmental Studies, University of Wisconsin–Superior, Superior, Wisconsin. pp. 51–52.

German, I.V. 1986. [Level of chromosome aberrations in workers coming in contact with hexachlorobutadiene during production.] Gig. Tr. Prof. Zabol. 5: 57–59 (in Russian).

German, I.V. 1988. Mutagenic activity of the pesticide hexachlorobutadiene. Tsitol. Genet. 22: 40–42.

Goldbach, R.W., H. Van Genderen and P. Leeuwangh. 1976. Hexachlorobutadiene residues in aquatic fauna from surface water fed by the river Rhine. Sci. Total Environ. 6: 31–40.

Govind, R., P.A. Flaherty and R.A. Dobbs. 1991. Fate and effects of semivolatile organic pollutants during anaerobic digestion of sludge. Water Res. 25: 547–556.

Gradiski, D., P. Duprat, J.L. Magadur and E. Fayein. 1975. Étude toxicologique experimentale de l'hexachlorobutadiene. Eur. J. Toxicol. 8: 180–187.

Graham, H. 1993. Personal communication, letter dated September 17, 1993. Water Resources Branch, Ontario Ministry of the Environment.

Gulko, A.G., N.I. Zimina and I.G. Shroit. 1964. Toxicological study of the insecticide hexachlorobutadiene. Vopr. Gig. Sanit. Ozdorovl. Vneshn. Sredy, Kishinev. Sb. [abstracted in Chem. Abstr. 62: 13757c (1965)].

Hardin, B., G.P. Bond, M.R. Sikov, F.D. Andrew, R.P. Beliles and R.W. Niemeier. 1981. Testing of selected workplace chemicals for teratogenic potential. Scand. J. Work- Environ. Health 7 (Suppl. 4): 66–75.

Harleman, J.H. and W. Seinen. 1979. Short-term toxicity and reproduction studies in rats with hexachloro-(1,3)-butadiene. Toxicol. Appl. Pharmacol. 47: 1–14.

Haworth, S., T. Lawlor, K. Mortelmans, W. Speck and E. Zeiger. 1983. *Salmonella* mutagenicity test results for 250 chemicals. Environ. Mutagen. 5 (Suppl. 1): 3–142.

Health Canada. 1994. Human health risk assessment for Priority Substances. *Canadian Environmental Protection Act.* Ottawa, Ontario.

Henschler, D. and W. Dekant. 1990. Nephrocarcinogenic xenobiotics. Toxicol. Lett. 53: 105–110.

Howard, P. 1991. Handbook of environmental fate and exposure data for organic chemicals. Lewis Publishers, London, U.K. pp. 360–369.

Howard, P., R. Boethling, W. Jarvis, W. Meylan and E. Michalenko. 1991. Handbook of environmental degradation rates. H. Lewis Publishers, Boca Raton, Florida.

IARC (International Agency for Research on Cancer). 1979. IARC monographs on the evaluation of the carcinogenic risk of chemicals to humans. Volume 20. Hexachlorobutadiene. World Health Organization, Lyon, France.

IPCS (International Programme on Chemical Safety). 1994. Environmental Health Criteria 156 — Hexachlorobutadiene. World Health Organization, Geneva, Switzerland.

Jaffe, D.R., C.D. Hassal and K. Brendel. 1983. *In vivo* and *in vitro* nephrotoxicity of the cysteine conjugate of hexachlorobutadiene. J. Toxicol. Environ. Health 11: 857–867.

Jobb, B., R. Hunsinger, O. Meresz and V. Taguchi. 1993. Removal of N-nitrosodimethylamine from the Ohsweken (Six Nations) water supply. Interim report, July 1993. Ontario Ministry of Environment and Energy.

Johnson, L.D. and J.C. Young. 1983. Inhibition of anaerobic digestion by organic priority pollutants. J. Water Pollut. Control Fed. 55(12): 1441–1449.

Jonker, D., R.A. Woutersen, P.J. van Bladeren, H.P. Til and V.J. Feron. 1993. Subacute (4-wk) toxicity of a combination of four nephrotoxicants in rats: compared with the toxicity of the individual compounds. Food Chem. Toxicol. 31: 125–136.

Juhnke, I. and D. Lüdemann. 1978. Results of the study of 200 chemical compounds on acute fish toxicity using the golden orfe test. Z. Wasser Abwasser Forsch. 11(5): 161–164.

Kauss, P.B. 1996. Personal communication, December 20, 1996. Ontario Ministry of Environment and Energy, Etobicoke, Ontario.

Kauss, P.B. 1997. Personal communication, May 5, 1997. Ontario Ministry of Environment and Energy, Etobicoke, Ontario.

Kauss, P.B. and Y.S. Hamdy. 1985. Biological monitoring of organochlorine contaminants in the St. Clair and Detroit rivers using introduced clams (*Elliptio complanatus*). J. Great Lakes Res. 11(3): 247–263.

Kendall, P.R.W. 1990. The quality of drinking water in Toronto: A review of tap water, bottled water and water treated by point-of-use device. Environmental Protection Office, Department of Public Health, City of Toronto.

Knie, J., A. Haelke, I. Juhnke and W. Schiller. 1983. Results of studies of chemical substances using four biotests. Dtsch. Gewaesserkd. Mitt. 27(3): 77–79.

Kociba, R.J., P.G. Gehring, C.G. Humniston and G.L. Sparschu. 1971. Toxicologic study of female rats administered hexachlorobutadiene or hexachlorobenzene for thirty days. The Dow Chemical Company, Midland, Michigan [cited in Kociba *et al.*, 1977b; IPCS, 1994].

Kociba, R.J., D.G. Keyes, G.C. Jersey, J.J. Ballard, D.A. Dittenber, J.F. Quast, C.E. Wade, C.G. Humiston and B.A. Schwetz. 1977a. Results of a two year chronic toxicity study with hexachlorobutadiene in rats. Am. Ind. Hyg. Assoc. J. 38: 589–602.

Kociba, R.J., B.A. Schwetz, D.G. Keyes, G.C Jersey, J.J. Ballard, D.A. Dittenber, J.F. Quast, C.E. Wade and C.G. Humiston. 1977b. Chronic and reproduction studies of hexachlorobutadiene in rats. Environ. Health Perspect. 21: 49–53.

Kotzias, D., W. Klein and F. Korte. 1975. [Analysis of residues of hexachlorobutadiene in foodstuff and poultry.] Chemosphere 4: 247–250 (in German).

Kusz, P., A. Andriysiak and Z. Pokorska. 1984. Gas chromatographic monitoring of the chlorolysis processes of some byproducts from vinyl chloride, allyl chloride and epichlorohydrin production. J. Chromatogr. 286: 287–291.

Laseter, J.L., C.K. Bartell, A.L. Laska, D.G. Holmquist and D.B. Condie. 1976. Ecological study of hexachlorobutadiene (HCBD). U.S. Environmental Protection Agency, Washington, D.C. (EPA-68-01-2689).

Laska, A.L., C.K. Bartell, D.B. Condie, J.W. Brown, R.L. Evans and J.L. Laseter. 1978. Acute and chronic effects of hexachlorobenzene and hexachlorobutadiene in red swamp crayfish (*Procambarus clarki*) and selected fish species. Toxicol. Appl. Pharmacol. 43(1): 1–12.

Leeuwangh, P., H. Bult and L. Schneiders. 1975. Toxicity of hexachlorobutadiene in aquatic organisms. Sublethal effects of toxic chemicals on aquatic animals. Proceedings of the Swedish–Netherlands Symposium, September 2–5, 1975. Elsevier, Amsterdam, The Netherlands. 167 pp.

L'Italien, S. 1996. Organic contaminants in Lake Ontario in 1992 and 1993: Protocols and data report. Ecosystem Health Division, Environmental Conservation Branch, Ontario Region, Environment Canada, Burlington, Ontario (EHD/ECD/-OR/96-01/I).

Lock, E.A. 1994. The role of mechanistic studies in understanding target organ toxicity. Arch. Toxicol. Suppl. 16: 151–160.

Lock, E.A. and J. Ishmael. 1979. Original investigations: the acute toxic effects of hexachloro-1:3-butadiene on the rat kidney. Arch. Toxicol. 43: 47–57.

Lock, E.A., J. Ishmael and J.B. Hook. 1984. Nephrotoxicity of hexachloro-1,3-butadiene in the mouse: The effect of age, sex, strain, monooxygenase modifiers, and the role of glutathione. Toxicol. Appl. Pharmacol. 72: 484–494.

Malins, D.C., M.M. Krahn, M.S. Nyers, L.D. Rhoses, D.W. Brown, C.A. Krone, B.B. McCain and S.I. Chan. 1985. Toxic chemicals in sediments and biota from a creosote-polluted harbor: relationships with hepatic neoplasms and other hepatic lesions in English sole (*Parophrys vetulus*). Carcinogenesis 6: 1463–1469 [cited in IPCS, 1994].

Manahan, S. 1992. Fundamentals of environmental chemistry. Lewis Publishers, Boca Raton, Florida.

Mayer, F.L. and M.R. Ellersieck. 1986. Manual of acute toxicity: Interpretation and data base for 410 chemicals and 66 species of freshwater animals. U.S. Fish Wildl. Serv. Resour. Publ. 160: 1–226.

McCarthy, R., G.M. Hawksworth and E.A. Lock. 1992. Subcellular localisation of C-S lyase activity in human renal cortex. 13th European Workshop on Drug Metabolism, September 21–25, 1992, Bergamo, Italy [cited in Lock, 1994].

McConnell, G., D.M. Ferguson and C.R. Pearson. 1975. Chlorinated hydrocarbons in the environment. Endeavor 34: 13–18.

Meek, M.E., R. Newhook, R.G. Liteplo and V.C. Armstrong. 1994. Approach to assessment of risk to human health for Priority Substances under the *Canadian Environmental Protection Act*. Environ. Carcinogen. Ecotoxicol. Rev. C12(2): 105–134.

Mes, J., D. Davis, D. Turton and W.F. Sun. 1986. Levels and trends of chlorinated hydrocarbon contaminants in the breast milk of Canadian women. Food Addit. Contam. 3: 313–322.

Ministers' Expert Advisory Panel. 1995. Report of the Ministers' Expert Advisory Panel on the second Priority Substances List under the *Canadian Environmental Protection Act* (CEPA). Government of Canada, Ottawa, Ontario. 26 pp.

Mudroch, A., R.J. Allan and S.R. Joshi. 1992. Geochemistry and organic contaminants in the sediments of Great Slave Lake, Northwest Territories, Canada. Arctic 45(1): 10–19.

Murzakaev, F.G. 1963. Toxicity data for hexachlorobutadiene and its intermediates. Farmakol. Toksikol. 26: 750 [abstracted in Chem. Abstr. 60: 13776b (1964)].

Nash, J.A., L.J. King, E.A. Lock and T. Green. 1984. The metabolism and disposition of hexachloro-1:3-butadiene in the rat and its relevance to nephrotoxicity. Toxicol. Appl. Pharmacol. 73: 124–137.

NIOSH (National Institute for Occupational Safety and Health). 1981. Tier II mutagenic screening of 13 NIOSH priority compounds: Individual compound report hexachloro-1,3-butadiene. Cincinnati, Ohio (PB83-152397).

NPRI (National Pollutant Release Inventory). 1994. Summary report 1994 national pollutant release inventory. Prepared by M. Nobert, Environment Canada (Cat. No. EN40-495\1-1994E; ISBN 0-662-24996-8).

NTP (National Toxicology Program). 1990. Final report on the peri-/postnatal evaluation of hexachloro-1,3-butadiene (HCBD) toxicity in CD rats. Research Triangle Park, North Carolina.

NTP (National Toxicology Program). 1991. Toxicity studies of hexachloro-1,3-butadiene in B6C3F$_1$ mice (feed studies). Research Triangle Park, North Carolina (NTP TOX 1; NIH Publication No. 91-3120).

Oliver, B.G. and K.L.E. Kaiser. 1986. Chlorinated organics in nearshore waters and tributaries of the St. Clair River. Water Pollut. Res. J. Can. 21(3): 344–350.

Oliver, B.G. and K.D. Nicol. 1982. Gas chromatographic determination of chlorobenzenes and other chlorinated hydrocarbons in environmental samples using fused silica capillary columns. Chromatographia 16: 336–340.

Oliver, B.G. and A.J. Niimi. 1983. Bioconcentration of chlorobenzenes from water by rainbow trout: correlations with partition coefficients and environmental residues. Environ. Sci. Technol. 17(5): 287–291.

Oliver, B.G. and C.W. Pugsley. 1986. Chlorinated contaminants in St. Clair River sediments. Water Pollut. Res. J. Can. 21(3): 368–379.

OME (Ontario Ministry of the Environment). 1991. St. Clair River MISA pilot site investigation. Volume II, Part II — Detailed technical findings. Municipal/Industrial Strategy for Abatement (MISA).

OME (Ontario Ministry of the Environment). 1992. Twelve-month monitoring data report — Organic Chemical Manufacturing Sector (Period covered: October 1, 1989 to July 31, 1991). Prepared by T.M. Tuszynski, Municipal/Industrial Strategy for Abatement (MISA) Section, Water Resources Branch, Ontario Ministry of the Environment, September 1992 (PIBS 2112, Log 92-2310-046).

OMEE (Ontario Ministry of Environment and Energy). 1996. Unpublished data from the Ontario Drinking Water Surveillance Program, provided by John McGrachan, Environmental Monitoring and Reporting Branch, Etobicoke, Ontario, May 2, 1996.

OME/MDNR (Ontario Ministry of the Environment/Michigan Department of Natural Resources). 1991. The St. Clair River Area of Concern. Environmental conditions and problem definitions: Remedial Action Plan Stage I. Submission to the International Joint Commission, December 20, 1991.

Pearson, C.R. and G. McConnell. 1975. Chlorinated C1 and C2 hydrocarbons in the marine environment. Proc. R. Soc. London 189(B): 305–332.

Reichert, D., T. Neudecker, U. Spengler and D. Henschler. 1983. Mutagenicity of dichloroacetylene and its degradation products trichloroacetyl chloride, trichloroacyloyl chloride and hexachlorobutadiene. Mutat. Res. 117: 21–29.

Reichert, D., T. Neudecker and S. Schutz. 1984. Mutagenicity of hexachlorobutadiene, perchlorobutenoic acid and perchlorobutenoic acid chloride. Mutat. Res. 137: 89–93.

Riopel, A. 1996. Personal communication, letter dated April 16, 1996. Unpublished data, Gouvernement du Québec, Ministère de l'Environnement et de la Faune, Direction des politiques du secteur municipal, Service de l'assainissement des eaux et du traitement des eaux de consommation.

Roederer, G., F. Brüggemann, H. Schäfer, K. Schöne, A. König and J. Steinhanses. 1989. Testung wassergefährdener stoffe als grundlage für wasser-qualitätsstandards. Testbericht, Fraunhofer-Institut für Umweltchemie und Ökotoxikologie, Schmallenberg, Switzerland. pp. 8–20.

Roldán-Arjona, T., M.D. García-Pedrajas, F.L. Luque-Romero, C. Hera and C. Pueyo. 1991. An association between mutagenicity of the Ara test of *Salmonella typhimurium* and carcinogenicity in rodents for 16 halogenated aliphatic hydrocarbons. Mutagenesis 6(3): 199–205.

Saillenfait, A.M., P. Bonnet, J.P. Guenier and J. de Ceaurriz. 1989. Inhalation teratology study on hexachloro-1,3-butadiene in rats. Toxicol. Lett. 47: 235–240.

Schrenk, D. and W. Dekant. 1989. Covalent binding of hexachlorobutadiene metabolites to renal and hepatic mitochondrial DNA. Carcinogenesis 10: 1139–1141.

Schwetz, B.A., F.A. Smith and C.G. Humiston. 1977. Results of a reproduction study in rats fed diets containing hexachlorobutadiene. Toxicol. Appl. Pharmacol. 42: 387–398.

Shen, T.T. 1982. Air quality assessment for land disposal of industrial wastes. Environ. Manage. 6: 297–305.

SIDS Manual. 1994. Screening Information Data Set (SIDS) Manual of the OECD Programme on the Co-operative Investigation of High Production Volume (HPV) Chemicals. MANUAL\9405.DOC/July 1994.

Slooff, W. 1979. Detection limits of a biological monitoring system based on fish respiration. Bull. Environ. Contam. Toxicol. 23(4–5): 517–523.

Stott, W.T., J.F. Quast and P.G. Watanabe. 1981. Differentiation of the mechanism of oncogenicity of 1,4-dioxane and 1,3-hexachlorobutadiene in the rat. Toxicol. Appl. Pharmacol. 60: 287–300.

Tabak, H.H., S.A. Quave, C.I. Mashni and E.F. Barth. 1981. Biodegradability studies with organic priority pollutant compounds. J. Water Pollut. Control Fed. 53(10): 1503–1518.

Theiss, J.C., G.D. Stoner, M.B. Shimkin and E.K. Weisburger. 1977. Test for carcinogenicity of organic contaminants of United States waters by pulmonary tumor response in strain A mice. Cancer Res. 37: 2717–2720.

Toxic Release Inventory. 1997. Toxic Chemical Release Inventory. National Toxicology Information Program, National Library of Medicine, Bethesda, Maryland.

U.S. EPA (United States Environmental Protection Agency). 1980. Ambient water quality criteria for hexachlorobutadiene (HCBD). Criteria and Standards Division, Washington, D.C. (EPA 440/5-80-053).

Vamvakas, S., F.J. Kordowich, W. Dekant, T. Neudecker and D. Henschler. 1988. Mutagenicity of hexachloro-1,3-butadiene and its S-conjugates in the Ames test — role of activation by the mercapturic acid pathway and its nephrocarcinogenicity. Carcinogenesis 9: 907–916.

Van Duuren, B.L., B.M. Goldschmid, G. Loewengart, A.C. Smith, S. Melchionne, I. Seldman and D. Roth. 1979. Carcinogenicity of halogenated olefinic and aliphatic hydrocarbons in mice. J. Natl. Cancer Inst. 63: 1433–1439.

Walbridge, C.T., J.T. Fiandt, G.L. Phipps and G.W. Holcombe. 1983. Acute toxicity of ten chlorinated aliphatic hydrocarbons to the fathead minnow (*Pimephales promelas*). Arch. Environ. Contam. Toxicol. 12(6): 661–666.

Webber, M.D. and C. Wang. 1995. Industrial organic compounds in selected Canadian soils. Can. J. Soil Sci. 75(4): 513–524.

Wild, D., S. Schutz and D. Reichert. 1986. Mutagenicity of the mercapturic acid and other S-containing derivatives of hexachloro-1,3-butadiene. Carcinogenesis 7: 431–434.

Yang, R.S.H. 1988. Hexachloro-1,3-butadiene: toxicology, metabolism, and mechanisms of toxicity. Rev. Environ. Contam. Toxicol. 101: 121–137.

Yang, R.S., K.M. Abdo and M.R. Elwell. 1989. Subchronic toxicity studies of hexachloro-1,3-butadiene (HCBD) in B6C3F1 mice by dietary incorporation. J. Environ. Pathol. Toxicol. Oncol. 9: 323–332.

Yip, G. 1976. Survey of hexachloro-1,3-butadiene in fish, eggs, milk and vegetables. J. Assoc. Off. Anal. Chem. 59: 559–561.

Zanette, M. 1996. Personal communication. Unpublished data provided July 16, 1996, by Greater Victoria Water District.

Zhu, J. 1997. Hexachlorobutadiene levels in samples collected during multimedia exposure pilot study. Memo to R. Beauchamp, Priority Substances Section, Environmental Health Directorate, Health Canada, dated April 10, 1997.

APPENDIX A: Search strategies employed for identification of relevant data

Environmental assessment

Relevant data were identified from existing review documents, published reference texts and on-line searches conducted between January and April 1996. The databases searched included the following: ASFA (Aquatic Sciences and Fisheries Abstracts, Cambridge Scientific Abstracts), BIOSIS (Biosciences Information Services), CAB (Commonwealth Agriculture Bureaux), CESARS (Chemical Evaluation Search and Retrieval System, Ontario Ministry of the Environment and Michigan Department of Natural Resources), CHRIS (Chemical Hazard Release Information System), Current Contents (Institute for Scientific Information), ELIAS (Environmental Library Integrated Automated System, Environment Canada Library), Enviroline (R.R. Bowker Publishing Co.), Environmental Abstracts, Environmental Bibliography (Environmental Studies Institute, International Academy at Santa Barbara), GEOREF (Geo Reference Information System, American Geological Institute), HSDB (Hazardous Substances Data Bank, U.S. National Library of Medicine), Life Sciences (Cambridge Scientific Abstracts), NTIS (National Technical Information Service, U.S. Department of Commerce), Pollution Abstracts (Cambridge Scientific Abstracts, U.S. National Library of Medicine), POLTOX (Cambridge Scientific Abstracts, U.S. National Library of Medicine), RTECS (Registry of Toxic Effects of Chemical Substances, U.S. National Institute for Occupational Safety and Health), Toxline (U.S. National Library of Medicine), TRI93 (Toxic Chemical Release Inventory, 1993, U.S. Environmental Protection Agency, Office of Toxic Substances), USEPA-ASTER (Assessment Tools for the Evaluation of Risk, U.S. Environmental Protection Agency), WASTEINFO (Waste Management Information Bureau of the American Energy Agency) and Water Resources Abstracts (U.S. Geological Survey, U.S. Department of the Interior).

A survey of Canadian industry was carried out under authority of Section 16 of CEPA (Environment Canada, 1997b). Companies were required to provide information on uses, releases, environmental concentrations, effects or other data on HCBD that were available to them if they met the trigger quantity of 1 kg of HCBD per year. Reveal Alert was used to maintain an ongoing record of the current scientific literature pertaining to the environmental effects of HCBD. Data obtained after November 30, 1997 were not considered in this assessment.

Human health assessment

Evaluations of other agencies such as the International Programme on Chemical Safety (IPCS, 1994) and the Agency for Toxic Substances and Disease Registry (ATSDR, 1994) were consulted and used to identify relevant data. Additional relevant data were identified through searches on the following databases in the fall of 1993: AQUAREF (Inland Waters Directorate, Environment Canada), CCRIS (Chemical Carcinogenesis Research Information System, U.S. National Cancer Institute), ChemID (U.S. National Library of Medicine; available on the Medical Literature Analysis and Retrieval System), CISTIMON (Canadian Institute for Scientific and

Technical Information list of monographs, National Research Council of Canada), DART (Developmental and Reproductive Toxicology, U.S. National Library of Medicine), ELIAS (Environmental Library Integrated Automated System, Environment Canada library), EMIC (Environmental Mutagen Information Center database, Oak Ridge National Laboratory), EMICBACK (backfile of EMIC), Enviroline (R.R. Bowker Publishing Co.), Environmental Bibliography (Environmental Studies Institute, International Academy at Santa Barbara), ETICBACK (backfile of Environmental Teratology Information Center database, U.S. Environmental Protection Agency and U.S. National Institute of Environmental Health Sciences), Food Science and Technology Abstracts, GENE-TOX (Genetic Toxicology, U.S. Environmental Protection Agency), HSDB (Hazardous Substances Data Bank, U.S. National Library of Medicine), IRIS (Integrated Risk Information System, U.S. Environmental Protection Agency), Microlog (Canadian Research Index, Government Publications, Micromedia Ltd.), Pollution Abstracts (Cambridge Scientific Abstracts, U.S. National Library of Medicine), RTECS (Registry of Toxic Effects of Chemical Substances, U.S. National Institute for Occupational Safety and Health) and Toxline (U.S. National Library of Medicine). Since these initial searches, the Canadian Research Index, Current Contents, Dialog, Medline, Toxline and Toxnet have been searched on a regular basis to identify recent articles. A general search of Internet web sites was performed in July 1996. Only data acquired prior to December 1996 were considered in the determination of whether HCBD is "toxic" to human health.